Interaction of backfilling techniques and MTA plugs with additives: Fracture strength and adaptation analyses

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The aims were to evaluate the effects of different additives on the adaptation of mineral trioxide aggregate (MTA) plugs before and after different backfilling techniques and analyze the interference of filling procedures on the fracture resistance of simulated immature teeth. The apical parts of 60 teeth were filled with MTA mixed with distilled water (DW), propylene glycol (PG) or calcium chloride (CaCl₂) and backfilled with cold lateral condensation (CLC) or warm vertical compaction (WVC). The specimens were subjected to adaptation analysis and fracture testing. CaCl₂ addition resulted in poor marginal adaptation, while PG addition significantly decreased the fracture values (p<0.05). WVC technique negatively affected the marginal adaptation of MTA-CaCl₂ plugs and decreased the fracture values of teeth with MTA-PG plugs (p<0.05). Mixing MTA with DW can be recommended as apical plug when the remaining root canal space is going to be filled with either CLC or WVC techniques.

Keywords: Apexification, Calcium silicate, Replica techniques, Root canal obturation

INTRODUCTION

Traumatic dental injuries can cause the loss of pulp vitality before development of the root is completed¹⁾. The resultant flaring root canals accompanied by thin immature dentinal walls render these teeth more prone to fracture¹⁾. Furthermore, these teeth occasionally have wide-open apices, which may be challenging to treat with conventional endodontic procedures¹⁾.

Historically, the treatment of an immature tooth with a necrotic pulp was performed with long-term calcium hydroxide placement inside the root canal system, with success rates ranging from $79-96\%^{2,3}$. However, this procedure has several drawbacks such as multiple visits, the need of patient compliance and the potential to further decrease the strength of the root⁴⁾. To overcome these concerns, an alternative treatment method, one-step apexification with mineral trioxide aggregate (MTA), has been proposed⁵⁾. This procedure can be completed in a single visit by applying orthograde placement of an artificial apical barrier, or plug comprised of MTA⁶. MTA is a calcium-silicate based material and possesses several desirable properties such as superior sealing ability, biocompatibility, and the ability to set in the presence of tissue fluids⁷. On the other hand, MTA presents some notable shortcomings such as difficult handling property and long setting time⁸⁾. To improve such properties of MTA, researchers have mixed it with different additives including propylene glycol (PG) and calcium chloride (CaCl₂)⁸⁻¹⁰⁾.

Although the immediate apexification can be provided by MTA, still there is a necessity of obturating the remaining canal space for sealing purposes and providing a reinforcement of the weak structure¹¹.

Following the apexification with MTA, backfilling techniques may affect the marginal adaptation of the apical plug. However, this issue has not been addressed in the literature. Furthermore, there is a lack of information regarding the fracture resistance of immature teeth backfilled with different techniques following the apexification with MTA including different additives. Therefore, the aims of the present study were to evaluate the marginal adaptation of MTA mixed with distilled water (DW), PG or CaCl₂ after backfilling procedures with cold lateral compaction (CLC) or warm vertical compaction (WVC) of gutta-percha and to test the fracture strength of these simulated immature teeth. The first null hypothesis of this study was that the additives to MTA has no significant influence on the marginal adaptation of the material and the fracture resistance of simulated immature teeth. The second null hypothesis was that the backfilling technique does not affect the marginal adaptation of MTA plugs with different additives and the fracture resistance of simulated immature teeth.

MATERIALS AND METHODS

Following the ethics committee approval (Ethics Board No: GO 16/334), 80 freshly extracted, human maxillary anterior teeth with similar dimensions were used in the study. The teeth were observed under magnification to confirm that there were no resorptions, extensive caries, cracks, or deformities. The crowns and 3 mm apical portions were removed with discs to obtain standard length of 13 mm for each sample and to simulate immature root apices similar to a previous study¹²⁾. Measurements of the samples were made using a

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digital caliper (Mitutoyo, Hampshire, UK). Ten samples were left without performing any additional procedure and served as negative control. The root canals of the remaining samples (n=70) were enlarged using ProTaper rotary files (Dentsply Maillefer, Ballaigues, Switzerland) up to F4 mounted on a torque-controlled reduction hand-piece (X-Smart, Dentsply Maillefer). For the simulation of immature apices, Peeso reamers (Dentsply Maillefer) between #2 and #6 were introduced in the root canals and a #6 Peeso reamer was allowed to protrude 1 mm beyond the apex to create an apical diameter of 1.70 mm. The root canals were irrigated using 2 mL of 5.25% sodium hypochlorite after each file and a final flush with 5 mL of 17% EDTA was made to remove the smear layer. Finally, the root canals were flushed with distilled water and dried using paper points (Dentsply Maillefer). At this point, 10 teeth were randomly selected for positive control group and remained enlarged and unfilled to simulate immature roots with thin walls. Throughout the experiments, the teeth were kept in moistened gauze.

To mimic immediate barrier formation, the apical 3 mm of each tooth was filled with MTA (ProRoot, Dentsply, Tulsa Dental, Tulsa, OK, USA) plugs using a hand plugger (SybronEndo, Kerr Endodontics, Orange, CA, USA) according to the groups below;

- Group DW (n=20): 1.0 g MTA powder was mixed with 0.35 mL DW.
- Group PG (n=20): 1.0 g MTA powder was mixed with 0.35 mL liquid composed of 20% PG (Emir Kimya, Ankara, Turkey)-80% DW by volume.
- Group $CaCl_2$ (n=20): 1.0 g MTA powder with 0.1 g $CaCl_2$ (Sisco Research Laboratories, Mumbai, India) was mixed with 0.35 mL DW.

Following the placement of the materials, each surface was gently cleaned with a moist gauze in order to clean the dentin surface from any material remnants. Digital periapical radiographs from the same angles were taken from each specimen to verify the density and the location of the apical plug. The samples were kept for 7 days at 37°C under 100% humidity.

Following the incubation period, each experimental group was divided into 2 subgroups according to the backfilling technique applied:

Groups DW-CLC, PG-CLC and CaCl₂-CLC: The remaining root canal space was backfilled using a root canal sealer (AH Plus, Dentsply, DeTrey, Konstanz, Germany) and gutta-percha (Dentsply) by CLC technique. In this technique, the sealer was mixed and inserted into the root canals using a lentulo spiral drill, and then a master cone was dipped in the sealer and introduced in the root canal until the working length. Then, accessory cones were inserted in the same manner and condensed using finger spreaders. Excess material was removed and vertical compaction was made using a cold plugger 1 mm below the orifice of the root canal.

Groups DW-WVC, PG-WVC and CaCl₂-WVC: The sealer was mixed and inserted into the remaining root canal space using a lentulo spiral drill. Backfilling was performed with the Elements[™]Free Obturation System (SybronEndo, Kerr Endodontics) at 200°C followed by compaction with a plugger.

Final radiographs were obtained to confirm uniform and dense obturation. The orifice of each root canal was sealed with a composite filling material (Filtek Z250, 3M ESPE, Seefeld, Germany). The teeth were stored at 37°C and 100% humidity for 1 week to allow the sealer setting.

Marginal adaptation analysis

Impressions of each air-dried root-ends were taken with vinylpolysiloxane precision impression material (Variotime, Heraeus Kulze, Hanau, Germany) before and after the backfillings and used as negative replicas to evaluate the effect of obturation techniques on the marginal integrity of the apical plugs according to the following 3-grade scale¹³;

Score 1: Material is well adapted, defined as follows:

- 1. Close marginal approximation of the filling material to the dentinal wall
- 2. No spacing defects present at the material-dentin interface in >70% of the circumference of the open apex (Figs. 1A and B)

Score 2: Material is moderately adapted, defined as follows:

- 1. Close marginal approximation of the filling material to the dentinal wall
- 2. Presence of spacing defects at the material-dentin interface in 30–60% of the circumference of the open apex (Fig. 1C)

Score 3: Root filling material is poorly adapted, defined as follows:

- 1. Poor marginal approximation of the filling material to the dentinal wall
- 2. Major voids and/or significant spacing defects at the material dentin interface in >60% of the circumference of the open apex (Fig. 1D)

Negative replicas were gold-sputtered for examination in a scanning electron microscope (Nova, NanoSEM 430, FEI, Hillsboro, OR, USA). Photomicrographs were taken at 130x magnification. Regions of special interest were photographed at 500× magnification (Fig. 2). The scoring was done by two independent endodontists in a blind manner. When a difference occurred in the scoring of an image, the two examiners reevaluated the image and discussed it until an agreement was obtained.

Fracture testing

Following marginal adaptation analysis, the specimens were perpendicularly embedded in self-curing acrylic resin poured in identically shaped cylinders^{11,14}, leaving a gap of 2 mm between the top of the acrylic and the cemento-enamel junction. A Universal Testing Machine

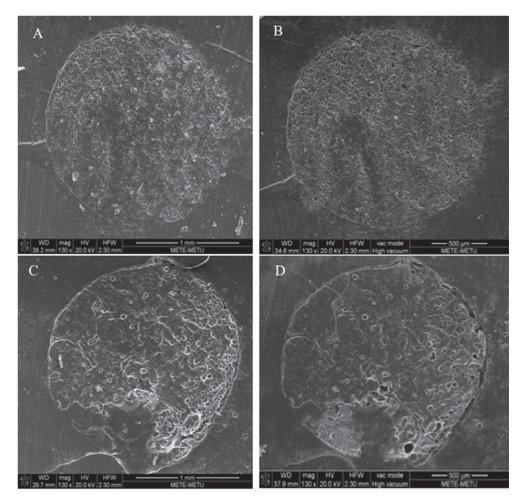


Fig. 1 Representative SEM images of the apical plugs.(A): Score 1 (before backfilling); (B): Score 1 (following backfilling); (C): Score 2 (before backfilling); (D): Score 3 (following backfilling).

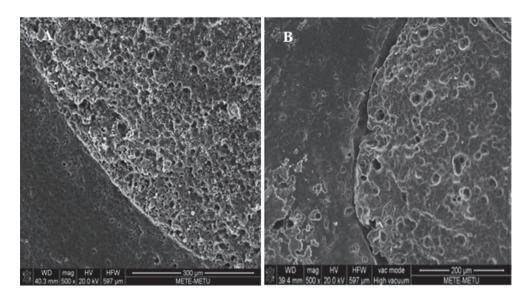


Fig. 2 Representative SEM images showing close marginal approximation in (A) and poor marginal approximation in (B) under 500× magnification.

(Lloyd LR 30K, Fareham, UK) was used to apply a compressive vertical loading. A 3-mm diameter circular stainless steel rod was affixed to the upper stage of the machine, while the acrylic blocks were mounted on the lower platform. The tip of the rod was centered over the canal orifice and a slowly increasing vertical force parallel to the long axis of the tooth was exerted (1 mm/min) until fracture occurred. The force when the fracture occurred was recorded in Newtons and fracture modes were analyzed as restorable and catastrophic (non-restorable). The fractures located on the cervical third of the root were classified as restorable while the fractures on the middle or apical third of the roots were catastrophic.

Statistical analysis

Kruskal Wallis and Mann Whitney U tests were used to compare score values before and after backfillings of intergroup. Wilcoxon Signed Ranks test was used to compare the adaptation scores within groups. Fracture resistance values were compared with univariate analysis of variance (ANOVA) with Bonferroni correction. In all comparisons, p value was set at 0.05.

RESULTS

Figure 3 shows the distribution of marginal adaptation scores of samples before and after the backfilling procedures. According to that, Group DW presented similar marginal adaptation to Group PG (p>0.05), while showed better adaptation than CaCl₂ before and after backfilling with both techniques (p<0.05). Group PG presented better marginal adaptation than Group CaCl₂ before both backfilling techniques and after CLC (p<0.05), while no significant difference was observed after WVC (p>0.05). The only significant difference regarding marginal adaptation change due to the backfilling procedure was seen in CaCl₂ after WVC (p<0.05). Regardless of the plug materials, there was no significant difference after backfilling with CLC and WVC in terms of marginal adaptation (p>0.05). The results regarding fracture testing are summarized in Table 1. The negative control group had the highest fracture resistance (p<0.05). The positive control group did not differ significantly from the experimental groups (p>0.05). Group PG-WVC showed significantly lower fracture resistance than Groups DW-WVC and PG-CLC (p<0.05). No significant difference was found among the other experimental groups in regards of fracture resistance (p>0.05). Regardless of the backfilling techniques, Group PG presented lower fracture resistance than Group DW (p<0.05). All samples were fractured in catastrophic way and most of the fractures

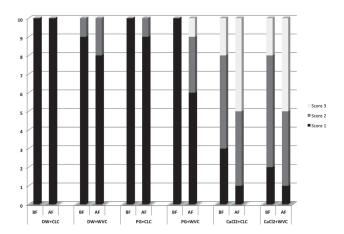


Fig. 3 Distribution of marginal adaptation scores of the experimental groups before and after backfilling. DW: distilled water; PG: propylene glycol; CaCl₂: calcium chloride; CLC: cold lateral compaction; WVC: warm vertical compaction.

Table 1 Fracture resistance values (Mean±Standard Deviation) in Newton and failure	e modes analysis of groups
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Groups	Fracture resistance	Failure modes		
		C/R	MD	$\mathbf{L}\mathbf{L}$
Negative control	583.40±93.15 °	С	2	8
Positive control	378.16 ± 60.35 bc	С	6	4
DW-CLC	447.68±83.92 ^b	С	4	6
DW-WVC	387.65 ± 68.72 ^b	С	3	7
PG-CLC	359.69 ± 68.93 ^b	С	2	8
PG-WVC	288.84±81.76°	С	6	4
$CaCl_2$ - CLC	362.35 ± 90.77 b	С	5	5
CaCl ₂ -WVC	350.69 ± 34.57 bc	С	2	8

* Different letters mean statistically significant differences, p < 0.05. C: catastrophic, R: restorable, MD: mesiodistal; LL: labiolingual

were labiolingually.

DISCUSSION

The use of MTA as an artificial barrier in the treatment of necrotic teeth with immature apices is a wellestablished technique¹⁵⁾. This treatment technique has been accomplished in several case reports by placing MTA plugs with a thickness of 3-5 mm and obturating the remaining root canal portion with cold lateral or warm vertical compaction techniques¹⁶⁻¹⁹⁾. Both obturation techniques may create different stresses and wedging effects inside the root canal system^{20,21}, thus may affect the marginal adaptation of apical plugs and also the fracture resistance of endodontically treated immature teeth. In the present study, we evaluated the effects of CLC and WVC techniques on the marginal adaptation and the fracture resistance of immature teeth obturated using MTA apical plugs with various additives.

Based on the results of the present study, PG and DW presented good marginal adaptation before and after both backfilling procedures. In a recent study, Duarte et al.²²⁾ found that the addition of PG, improved the flowability and the handling of MTA and they proposed 80% DW-20% PG as the optimum ratio. Also, in another study the same ratio was found to improve the push out bond strength of MTA⁸). Based on these studies, we decided to use this ratio for preparing PG in the present study. It was suggested that the addition of PG to MTA improves the consistency with more cohesiveness leading to easy manipulation and placement of the material into the defect site⁸⁾. Although better handling property was obtained with PG compared to DW, no significant difference was found in their marginal adaptation to dentin in the present study. In regards to this finding, we can recommend the use of PG as an apexification material; however further research is necessary to evaluate the other properties of this mixture. In several studies, it was reported that the addition of $CaCl_2$ accelerates the setting time of MTA^{9,10}). Since the long setting time is one of the main disadvantages of MTA, the use of CaCl₂ as an apexification material was considered as a reasonable choice in the present study. According to our results, CaCl₂ presented poor marginal adaptation compared to PG and DW. Moreover, the marginal adaptation of MTA apical plug was only affected by the obturation technique when only mixed with CaCl₂. In a recent study, the addition of CaCl₂ was associated with a negative influence on the push-out bond strength of MTA to dentin⁹⁾. The bond strength is responsible for resistance against procedural forces such as forces applied during the obturation of the root canal system⁸⁾. It is probable that lower bond strength of $CaCl_2$ to dentin⁹, led to a negatively influenced marginal adaptation after the backfilling procedure. Furthermore, a previous study showed that the compressive strength of MTA obtained with the addition of CaCl₂ was lower than MTA with DW¹⁰). Since compressive strength is a property that

indicates how a material bears under pressure, it can be another explanation why the material showed poor adaptation after the backfilling procedure. The addition of CaCl₂ accelerates the setting of MTA and thus reduces the incorporation of water²³⁾. The changed powderliquid ratio may be responsible from poor marginal adaptation of MTA-CaCl₂. Another interesting finding of the present study is that the adaptation of CaCl₂ was affected by only WVC technique, while no significant difference was occurred after CLC technique. The stresses occurred during lateral or vertical compaction is a controversial topic in literature. Gimlin et al.24) compared the lateral and vertical compaction forces using a mathematical model and found that lateral compaction was more likely to produce undesirable stress concentration than vertical compaction of guttapercha. On the other hand, a finite element analysis study showed that comparable magnitudes of stresses were occurred in dentin during both obturation techniques²⁵⁾. In the present study, only $CaCl_2$ group showed altered marginal adaptation after WVC technique and this could be related to the direct forces applied by the plugger on the material. According to the literature, the wedging effect of a spreader during lateral compaction and the force application by a plugger during vertical compaction are both risk factors for stress concentration at localized areas²⁰. However, it is noteworthy to mention that the aforementioned studies represent the teeth with closed apices. Further research is necessary to evaluate the stress concentration and distribution during different obturation techniques in immature teeth with wide open apices.

For evaluation of the marginal adaptation, we performed SEM examination, which is a commonly used method in laboratory studies owing to its ability to provide high magnification and good resolution²⁶⁻²⁸⁾. Importantly, the process of SEM preparation may affect the results because of the high vacuum evaporation and dehydration of the coating process of biological samples that can cause development of artifacts such as cracks in hard tissues and separation or lifting of the filling materials from the surrounding tooth structure²⁶. Furthermore, it is impossible to examine the same specimen with SEM before and after experimental procedures due to the coating process. To overcome these problems, we applied a replication technique by obtaining negative replicas of the original specimens using impression materials. Owing to this technique, we could examine the marginal adaptation of the same specimen before and after experimental procedures without any harm to the original specimens. In a previous study that evaluated quality of replication technique, low-viscosity silicones were found to be excellent materials for making impressions of tooth surfaces for SEM evaluation²⁹. According to the quality and the resolution of the SEM images obtained in the present study, the use of negative replicas can be recommended for future studies especially when before and after procedure images are necessary.

Apexification treatment by using MTA plugs is

highly successful^{16,30} and possesses several advantages such as shorter treatment time, creation of a good apical seal and an apical barrier on which obturation material might be condensed³⁰⁾. Despite these advantages, immature teeth are susceptible to fracture due to their thin and weak root canal walls⁴⁾. Therefore, it is essential to select a material or an obturation technique that would substantially exert a reinforcing effect. In the present study, strengthening capacity of MTA plugs with different additives and obturation techniques were tested by evaluating the fracture resistance of the teeth. For this analysis, we chose teeth with similar dimensions, performed decoronization prior to experimentation to standardize the samples and applied only vertical load along the long axis of the root to obtain uniform distribution of the force in each sample. Thus, the results obtained do not reflect the clinical situation directly, but can provide a relative comparison among the plug materials and obturation techniques. Within the limitations of this study, results have shown that only the addition of PG to MTA caused significant differences in fracture strength values. The lower fracture strength values obtained by the DW-PG group can be related to the moisture alteration in dentin tissue. It is well known that MTA requires water to initiate the hydration reaction and the setting process^{22,31}. According to a recent study, the use of PG reduced the amount of water available for hydration and thus, the addition of PG increased the setting time of MTA²²⁾. Therefore, the addition of PG to MTA may affect the water content of dentin negatively during the setting process, leading the tissues more susceptible to fracture. Furthermore, the force created by a plugger during vertical compaction may cause increased stress concentration at localized areas²⁰⁾ and could be associated with the further decrease in the fracture susceptibility of PG-WVC group.

CONCLUSION

Based on the results of this *in vitro* study, the both null hypotheses were rejected. The addition of $CaCl_2$ negatively influenced the marginal adaptation of MTA while the addition of PG to MTA caused significant decrease in the fracture resistance. The WVC technique caused deterioration in the marginal adaptation of MTA mixed with $CaCl_2$ and resulted in a significant decrease in the fracture resistance of teeth plugged with MTA/ PG mixture. The use of DW as an additive for preparing MTA apical plug in immature teeth can be recommended when the remaining root canal space is going to be filled with either CLC or WVC techniques.

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