



## Apical Revascularization of Necrotic Young Permanent Teeth by Different Methods

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### KEYWORDS

*Biodentine, MTA,  
Immature, Permanent teeth,  
Revascularization*

### ABSTRACT

**Aim:** to evaluate effect of Biodentine and MTA on the root length, root dentin thickness and apical diameter after revascularization in necrotic immature permanent teeth. **Subjects and methods:** 20 upper anterior teeth showed immature roots with an open apex, ranging in age from 9 to 12 years. Revascularization treatment was done, which was started with disinfection of the canals (sodium hypochlorite irrigation, followed by triple antibiotic paste for 2- 3 weeks). Next, the triple antibiotic paste was removed by irrigation with saline, The teeth were divided into two groups 10 each Group A: revitalization using blood clot scaffold only, and Group B: revitalization using blood clot and platelet rich fibrin (PRF). Each group was subdivided into two subgroups according to the material placed over the root canal orifice and a coronal seal of glass ionomer. Follow-up was done for 6 months. Standardized radiographs were analyzed for the peri-radicular healing and apical closure. **Results:** There was significant decrease in the mean apical diameter along the follow up periods. No significant differences were shown between all groups. **Conclusion:** Clinical and radiographic evidence showed, revascularization procedure could be an alternative treatment in immature nonvital teeth. In addition, placing Biodentine and MTA cement provided a good seal and favorable outcomes.

### INTRODUCTION

Traumatic injuries that cause fracture of the anterior teeth in children and adolescent are common these days. This type of injury can often cause a loss of pulp vitality and arrest to the root development of the affected teeth. Treating these teeth with apical infection and incomplete root formation with an open apex is a major challenge for dentist<sup>(1)</sup>.

While in most organs, the infection is treated by antibiotics, the dental pulp infection depends not only on controlling the infection locally, but also on restoring dental pulp vitality<sup>(2)</sup>. Although the use of local antimicrobial therapy alone, is effective in controlling pulp infections, it fails to restore growth of the tooth roots<sup>(3)</sup>.

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Apexification is the traditional method of treating immature permanent teeth with necrotic pulp to induce a calcified barrier in a root with an open apex or the continued apical closure of an incompletely formed root<sup>(4)</sup>. The apexification, is the use of calcium hydroxide to fill the root canals as an intracanal medicament because of its potential as an antimicrobial and root stimulation to complete growth towards the closure of the radical ends. The drawback of this approach is not only that it needs lots of visits and long times before the roots are filled<sup>(5)</sup>.

Conventional endodontic therapy using the traditional chemical mechanical tools and sodium hypochlorite has been shown to be ineffective in achieving appropriate goals (cleaning and disinfection) of the entire dentin wall, especially at the opened apex. But it also includes that, the walls of the root canals remain thin and continued root formation might not occur and such teeth after root canal treatment are prone to fracture<sup>(6)</sup>.

An innovation in the endodontic field is the use of the generation of a functional pulp-dentin complex as an alternative technique to replace traditional apexification in an attempt to stimulate further root growth and thickening of the dentinal walls at an immature non vital tooth<sup>(7-8)</sup>.

Since 1950 and 1960, scientific research has focused on trying to revascularization to the injured pulp of immature permanent teeth as a result of trauma<sup>(9)</sup>. It has been observed that, re-implanted teeth after trauma can regain their vitality despite the injury. There are many studies focused on the possibility of revascularization of the contaminated root canals if their environment is improved with disinfection. The efficacy of antibiotic paste in disinfecting the immature teeth of dogs with apical periodontitis was determined Wendley et al<sup>(10)</sup>. Wendley suggested that revascularization of necrotic immature permanent teeth can be predicted if three conditions were met: disinfecting the canals, placing a scaffold matrix to allow tissue to grow, and tight coral seal against bacteria.

In recent years, revascularization procedures have emerged as a new treatment method for immature necrotic permanent teeth. This new treatment involving, induction of intracanal blood clot formation, which has been previously disinfected by the introduction of stem cells from the apical area<sup>(11)</sup>. Currently, there are two ways to regenerate tissue in the pulp. The first is revascularization, in which new pulp tissue is expected to grow in the root canal from the remaining tissue in the root canal<sup>(12)</sup>. The second approach is to replace the injured pulp with healthy tissue, which can revitalize the teeth and restore the dentin formation process<sup>(13)</sup>.

With this treatment, immature tooth can continue to complete the root growth, strengthening the root wall by increasing the thickness of the dentin which prevent root fractures, apical closure, and allowing for complete elimination of the periapical bone lesion<sup>(14)</sup>. There are many reports of revascularization not only in single rooted teeth but also in multirooted teeth<sup>(15)</sup>.

Platelet-rich plasma (PRP) has been reported for bone augmentation<sup>(16)</sup>, and can be an ideal matrix in regenerative endodontic therapies because it meets many of the appropriate matrix properties<sup>(17)</sup>. PRP is a self-absorbing substance that is easily reabsorbed and rich in growth factors found in the alpha granules of platelets<sup>(18,19)</sup>. In addition, PRP makes a 3D matrix that supports adhesion and cell migration, helping proliferation of stem cells to induce repair and/or tissue regeneration.

A bioactive material has been defined as a material that has an impact on or stimulates response to living tissues, organisms, or cells, such as inducing the formation of hydroxyapatite. The ideal properties of bioactive materials are bactericides, bacteriostatic, bactericidal, sterile, stimulate the formation of reparative dentine, and maintain the vitality of the pulp. Various bioactive materials have been studied, including calcium hydroxide, mineral trioxide aggregate (MTA), calcium-enriched mixture (CEM), Biodentine and inert materials (isobutyl



cyanoacrylate and tricalcium phosphate ceramic) were studied<sup>(20,21)</sup>.

MTA has been successfully used in the clinical endodontic field for a long time, with acceptable biological compatibility and good in vivo and in vitro biologic behavior<sup>(22)</sup>. Multiple clinical studies have shown successful regenerative endodontic therapy outcomes using MTA. MTA can be used to seal the root canal, providing an environment conducive to tissue regeneration and repair<sup>(23)</sup>. However, it has showed some drawbacks, such as a change in color.

Biodentine is new a tooth-colored calcium silicate-based cement of the same or higher properties as MTA, developed as a bioactive dentin substitute and having mechanical properties like dentin properties. It has mechanical properties similar to sound dentin and can be replaced both in the crown and at the root, without any initial conditioning of mineral tissue. Thus, Biodentine saves teeth by preserving the pulp and promoting healing of the pulp as well as eliminating the need for root canal therapy in most cases.<sup>(24)</sup>

Several studies compared the performance of Biodentine and MTA and stated that, performance was equal to or superior to that of MTA. In human teeth, there were similar clinical, radiological, and histological effects after covering the pulp directly with MTA and Biodentine<sup>(25)</sup>. Moreover, there was no significant difference in cell viabilities, inflammatory response, and odontoblastic differentiation<sup>(26)</sup> between Biodentine and MTA in human gingival fibroblasts and human dental pulp cells<sup>(27)</sup> suggesting that Biodentine could be a good alternative pulp capping agent. The purpose of this study was to evaluate effect of Biodentine and MTA on apical diameter after revascularization in necrotic immature permanent teeth.

Therefore, this study was designed to evaluate and compare the effect of Biodentine and MTA on apical diameter after revascularization in necrotic

immature permanent teeth. Revascularization achieved through these two methods, a conventional blood clot and Platelet-rich plasma (PRP).

## MATERIALS AND METHODS

### Study design

Twenty healthy children with upper anterior teeth that had necrotic pulp due to carious or traumatic exposure, and they were complaining of pain and/or mucosal swelling with signs and/or symptoms of periapical pathology and immature roots with open apex selected from patients ages 9 to 12, attending the Department of Pediatric Dentistry, Zagazig University Faculty of Oral and Dental Surgery.

### Inclusion criteria<sup>(28)</sup>:

- An immature open apex of at least 1.1 mm, either in the form of blunderbuss or a wide canal with parallel walls.
- An existing radiographic periapical pathosis.
- Cooperative child.

### Exclusion criteria:

- Children who were taken systemic antibiotics.
- Children with chronic diseases or root fractures.

An informed consent signed by the parent / guardian for the study was obtained after full explanation of treatment procedures, risks, benefits, and alternative treatments in case of unresponsiveness to treatment.

### First visit:

Clinical and radiological evaluation was performed to determine the pre-operative condition of teeth, in relation to tooth mobility, presence or absence of sinus tract, presence or absence of preapical pathology, pocket depth, and percussion testing. The Electric Pulp test was performed on the affected tooth and the corresponding tooth as a record.

Standard preoperative digital radiograph was taken using the standardized paralleling technique through Film Locator System (RinnXcp) (Rinn Corporation Elgin, Illinois, USA), VISTA ray charged coupled device (CCD) system (Vista Ray sensor 7.1, Durr Dental, Germany) and X-ray machine (Fona XDC, ViaGalilei, Assago, Italy). The VISTA ray CCD is a mini-digital X-ray system that takes X-rays directly on the side of a chair with an immediate image display. The active dimensions of the CCD sensor were 20x30mm, the size of 19x19 microns with a total pixel count of 1,659,000 pixels and DBSWIN 5.3.1. (Fig 1)



Fig. (1) A photograph showing paralleling technique.

Under rubber dam isolation, the access was prepared on all teeth with round diamond bur No. 2 and a high-speed hand piece. Minimal instrumentation was done using the K-file size #20 that was entered into the canal to determine the working length. A child should not have any pain during the procedure, which confirmed the diagnosis of pulp necrosis<sup>(29)</sup>. and root canals were irrigated using 10 ml of 2.5% of NaOCl to remove the necrotic tissues. Canals were then dried with paper points. Mixture of triple antibiotic in equal ratios of ciprofloxacin, metronidazole, and monocycline was ground and mixed with sterile saline solution to get a thick creamy paste. This antibiotic baste was placed into the canals using amalgam carrier and packed with large endodontic pluggers. Access cavities were sealed by Cavite (3 ESPE, Germany).

## Second visit:

Patients were recalled two weeks later, and the teeth were tested for sensitivity to palpation and percussion. If the tooth was free of symptoms, under the isolation of the rubber dam, the antibiotics baste were removed from the canals by using 2.5% sodium hypochlorite and normal saline irrigation. The canals dried again and one of the following procedures was carried out<sup>(30)</sup>.

## Groups:

The teeth were divided into two groups 10 each:

Group A: revascularization using blood clot scaffold only.

Group B: revascularization using blood clot and platelet rich fibrin (PRF).

## Group A:

A No. 40 K-file was introduced to the apical tissue beyond the foramen to induce bleeding within the canal. Bleeding was controlled at 3 mm below the cemento-enamel junction (Fig 2). A blood clot formed in the canal around 15 minutes after stimulation. Bleeding stimulation and blood clot stabilization in the root canal system via over instrumentation was completed by a good coronal seal.



Fig. (2) A photograph show induction of bleeding within the canal. of bleeding within the canal

**Group B:**

Instead of a blood clot alone, PRF membranes was introduced into the canal with cotton pliers or soaked on a collagen sponge and carried to the apical portion with a plugger 1 mm over length and a slightly lower to the level of CEJ and completed by a good coronal seal<sup>(31)</sup>. Each group was subdivided into two subgroups (1&2) according to the material placed over the root canal orifice.

**Subgroups A<sub>1</sub>&B<sub>1</sub>:** MTA (Dentsply Tulsa Dental, Johnson City, TN, USA) was mixed according to its manufacturing instructions and placed over the blood clot carefully in A1 group and directly on top of EPRF clot in B1 group. MTA was covered by wet cotton granule for 15 minutes for complete setting. Cavit temporarily filling was placed over the cotton pellet.

**Subgroups A<sub>2</sub>&B<sub>2</sub>:** Also, according to instructions from the manufacturer, tri calcium silicate cement (Biodentine) (Septodont Dental Ltda., Pomerode, Santa Catarina, Brazil) is mixed and placed just above the blood clot on top of the EPRF clot. Biodentin was left for 6 minutes to complete setting, followed by a moist cotton pellet. Cavit was placed temporarily over the cotton pellet (Fig 3).



Fig. (3) A photograph show placing the bioactive material (MTA)

After 72 hours, permanent restoration was placed with composite resin. Patients were recalled after 2 weeks, 3 and 6 months for postoperative follow-up. For teeth with persistent infection, or where the canal cannot be dried, the triple antibiotics mixture dressing was repeated until no symptom or exudation was present. On the other hand, if the patient or the parents have any complaint (pain and/or swelling) or became worried, then the revascularization procedure was discontinued, and apexification was performed.

**Platelet-rich fibrin preparation:**

Five mL of whole blood was collected from patient's median cubital vein. The blood sample was subjected to centrifugation at 2400 rpm for 12min to prepare PRF (Choukroun's method)<sup>(31)</sup>

**Follow-up:**

At each follow up visit, the patient was evaluated clinically and radiologically.

**Clinical assessment:**

- History and clinical examination:
  1. Presence or absence of pain (spontaneous, stimulant, or rhythm)
  2. Presence or absence of swelling or sinus duct.
  3. Electric pulp testing (EPT)
- Treatment was considered clinically successful on the following criteria:
  - a. No signs or symptoms of the treated teeth.
  - b. + response to vitality test using Electric pulp testing (EPT)

**Radiographic examination:**

- The measurements were performed using the Image-J analysis software (Image-J analysis software v1.44 National Institute of Health, USA) by another researcher who was blind to the group being studied to avoid bias.

- i. Change in root length.
- ii. Change in root thickness.
- iii. Change in apical diameter.

#### i. Change in root length:

A measurement scale in Image-J software was set by measuring a dimension known as the active sensor width in millimeters (20 mm) to its radiometric dimension in pixels, so the scale was calculated as the number of pixels measured per mm length.

Root lengths were measured as a straight line from the cement-enamel junction to the top of the radiographic apex in millimeters (Figure 4). The root length was measured Pre- and post-operative and, the difference in length was calculated, and then the percentage of increase in length was calculated as follows:

$$\text{Percent increase in length} = \frac{\text{postoperative length} - \text{preoperative length}}{\text{preoperative length}} \times 100$$

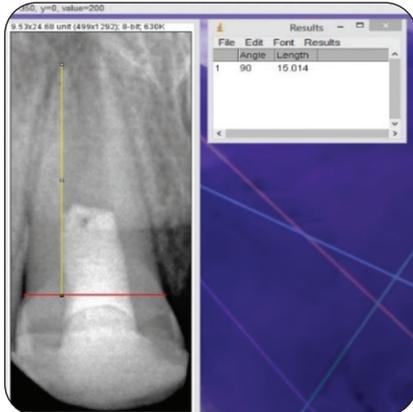


Fig. (4) Digital measurement of root length using image-J software.

#### ii. Change in root dentin thickness:

Using a preset measurement scale, the root thickness and pulp width were measured at the same fixed level in millimeters (Figure 5). Root dentine thickness was measured by subtracting the pulp space from the entire root thickness. Pre- and

post-operative measurements were performed at the same fixed level. The difference in thickness was calculated and the percentage increase in dentin thickness was calculated as follows:

$$\text{Dentin thickness} = \text{root thickness} - \text{pulp width}$$

$$\text{Percentage increase in thickness} = \frac{\text{postoperative thickness} - \text{preoperative thickness}}{\text{preoperative thickness}} \times 100$$

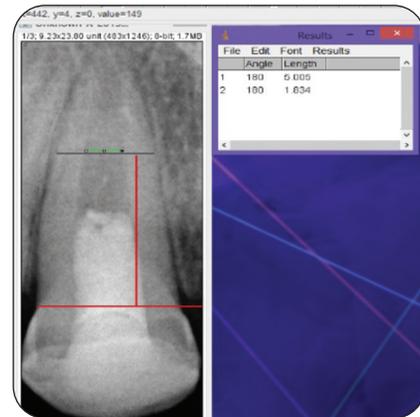


Fig. (5) Digital measurement of root dentin thickness using image-J software.

#### iii. Change in apical diameter:

Using the preset measurement scale, the diameter of the apical foramen was measured in millimeters. Pre- and post-operation measurements were made, and the variance was calculated in country diameter. The percentage closure on demand was calculated as follows:

Using the preset measurement scale, the diameter of the apical foramen had been measured in millimeters (Fig. 6). Measurements had been done pre and post operatively and the difference in apical diameter had been calculated. Percentage of apical closure had been calculated as follows:

$$\text{Percentage of apical closure} = \frac{\text{preoperative apical diameter} - \text{post operative apical diameter}}{\text{preoperative apical diameter}} \times 100$$

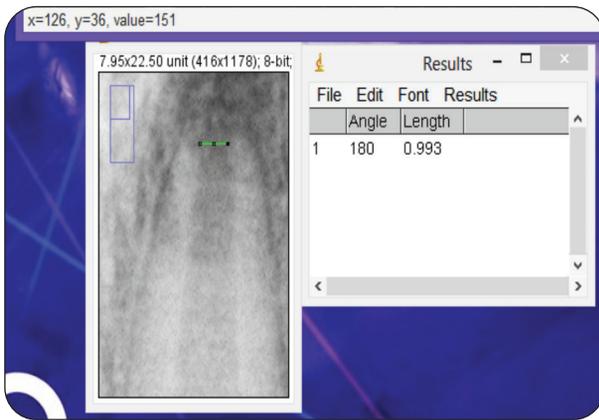


Fig. (6) Screen shot showing digital measurement of apical diameter using image J software.

### Treatment was considered radiographically successful on the following criteria:

Evidence of periapical healing, increased root length, and increased thickness of the root canal wall.

### Statistical analysis

All the study results were collected, tabulated, and expressed as mean  $\pm$  standard deviation (SD) and analyzed using (ANOVA) test followed by the LSD post hoc test with the use of the SPSS 17.0 software program (SPSS, Chicago, IL, USA).

A t test was used to compare the two groups. The results were considered statistically significant at  $\leq 0.5$ .

## RESULTS

This study was conducted to evaluate clinically the effect of Biodentine and MTA on apical diameter after revascularization in necrotic immature permanent teeth using blood clot and PRF.

### A) Clinical outcome:

**Group A<sub>1&2</sub>: (MTA & Biodentine):** Nine cases were evaluated, 4 MTA, 5 Biodentine and 1 from A1 was withdrawn due to incomplete follow-up.

There were no unfavorable signs or symptoms observed over the total follow up period. No sensitivity to percussion and palpation and palpation, also there were no swelling, erythema or sinus tracts. There were no reliable responses to electric pulp test.

**Group B<sub>1&2</sub>: (MTA & Biodentine):** (9 cases, 5 MTA and 4 Biodentine)

One case in B1(MTA) showed slight immediate pain, subsided within few hours. The remaining 8 cases didn't show any unfavorable signs or symptoms over the total follow up period. No sensitivity to percussion and palpation, also there were no swelling, erythema, or sinus tracts as well. There were no reliable responses to electric pulp test.

### B) Radiographic changes:

In all cases periapical radiolucency had disappeared, the overall radiographic evaluation of the two groups over the follow up periods is presented in (Fig 7&8)

#### i. Change in root length:

**Table (1)** shows the mean change in root length and percentage of change in each group at each interval.

#### Group A1&B1(MTA)

After 3 months, the mean increase in root length in group A<sub>1</sub>, was  $0.35 \pm 0.2$  mm representing 2.9%. while in B<sub>1</sub>, was  $0.37 \pm 0.22$  mm representing 3.3%. After 6 months, the mean increase in length was  $0.79 \pm 0.4$  mm representing 7%. in (group A<sub>1</sub>), while in group B1,  $0.81 \pm 0.39$  mm representing 7.01%.

#### Group A2&B2(Biodentine)

After 3 months, the mean increase in length in group A<sub>2</sub>, was  $0.45 \pm 0.2$  mm representing 2.89% while in B<sub>2</sub>, was  $0.42 \pm 0.19$  mm representing

3.27%. After 6 months, the mean increase in length was  $0.9 \pm 0.27$  mm representing 6.7% in A<sub>2</sub> group and  $0.88 \pm 0.26$  mm representing 6.68% in B<sub>2</sub> group.

Statistical analysis showed that there was increase in the root length without statistically significant difference between the two groups through the whole follow up period.

Table (2) shows the mean changes in root length and percentage of change at different time periods within each group. Regarding to the evaluation period at each group, there was significant increase in length at the MTA group.

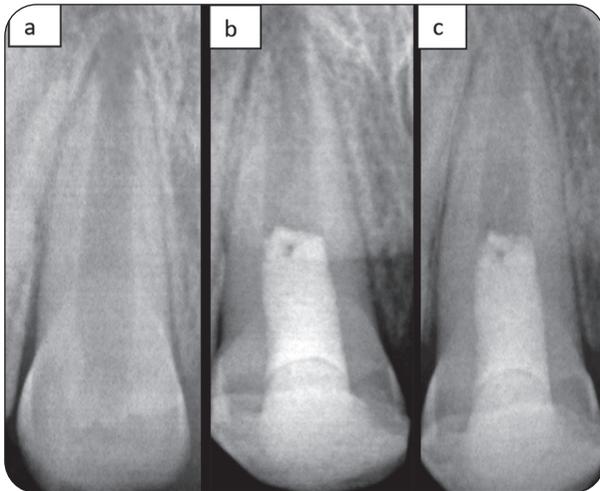


Fig. (7) Representative case in group A<sub>1</sub> (Blood clot & MTA group)  
Preoperative radiograph  
3 months post-operative radiograph  
6 months post-operative radiograph

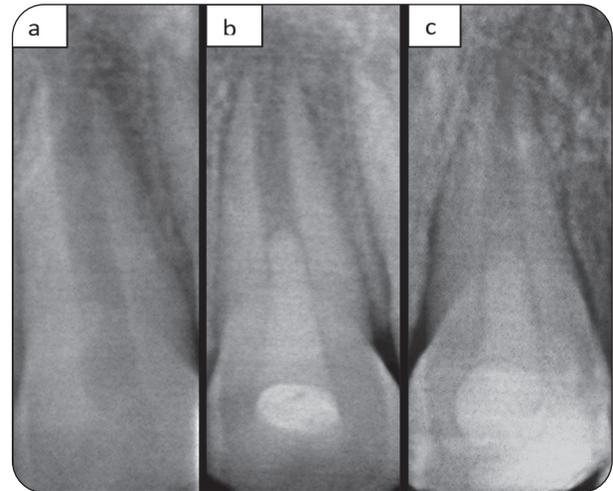


Fig. (8) Representative case in group B<sub>2</sub> (PRF & Biodentine group)  
Preoperative radiograph  
3 months post-operative radiograph  
6 months post-operative radiograph

**Table (1)** A comparison between the change in root length and the percentage change in the two groups in each period:

Time	Group A (Blood clot) Mean in mm $\pm$ SD (%)		Group B (RPF) Mean in mm $\pm$ SD (%)		P- value
	A <sub>1</sub> MTA	A <sub>2</sub> Biodentine	B <sub>1</sub> (MTA)	B <sub>2</sub> Biodentine	
3 months	$0.35 \pm 0.2$ (2.9%)	$0.45 \pm 0.2$ (3.3%)	$0.37 \pm 0.22$ (2.89%)	$0.42 \pm 0.19$ (3.27%)	0.327
6 months	$0.79 \pm 0.4$ (7%)	$0.9 \pm 0.27$ (6.7%)	$0.81 \pm 0.39$ (7.01%)	$0.88 \pm 0.26$ (6.68%)	0.535

\* The mean difference is significant at ( $P \leq 0.05$ )



**Table (2)** A comparison of the change in mean length and the percentage change in length in a different evaluation period within each group.

Time \ Group	3 months		6 months		P-value
	1(MTA)	2(Biodentine)	1(MTA)	2(Biodentine)	
Group A	0.35±0.2 (2.9%)	0.45±0.2 (3.3%)	0.79±0.4 (7%)	0.81±0.39(7.01%)	0.001**
Group B	0.37±0.22(2.89%)	0.42±0.19(3.27%)	0.9±0.27 (6.7%)	0.88±0.26(6.68%)	0.001**

\* is significant at the 0.05 level of significances ( $P \leq 0.05$ ),

\*\* is highly significant at the 0.001 level of significance ( $P \leq 0.001$ )

## ii. Change in dentine thickness

Table (3) shows the mean change in root dentin thickness and percentage of change in each group at each interval.

### Group A1&B1(MTA)

After 3 month the mean increase in root dentine thickness was  $0.06 \pm 0.02$  mm representing 3.2% in group A<sub>1</sub> while it was  $0.07 \pm 0.021$  mm representing 3.19% in B<sub>1</sub> group. After 6 months, the mean increase in root dentin thickness in A<sub>1</sub> group was  $0.12 \pm 0.02$  mm representing 6.36% and  $0.11 \pm 0.03$  mm representing 6.33% in group B<sub>1</sub>.

### Group A2&B2(Biodentine)

After 3 month the mean increase in root dentine thickness was  $0.11 \pm 0.02$  mm respectively 5.5% in A<sub>2</sub> group while it was  $0.10 \pm 0.01$  mm representing

5.52% in B<sub>2</sub> group. After 6 months, the mean increase in root dentin thickness was  $0.18 \pm 0.04$  mm representing 8.9% in A<sub>2</sub> group and  $0.19 \pm 0.04$  mm representing 8.97% in B<sub>2</sub> group.

Statistical analysis showed that there was highly statistically significant increase in the mean root dentin thickness at Biodentine group than MTA group through all follow up period.

Table (4) shows the mean change in root dentin thickness and percentage of change at different time periods within each group.

Regarding to the evaluation period at each group, there was significant increase in root dentin thickness at MTA group along the follow up periods. As well as there was significant increase in root dentin thickness at Biodentine group along the follow up periods.

**Table (3)** A comparison between the change in root dentition thickness and percentage of change in the two groups at each interval:

Time \ Group	Group A (Blood clot) Mean in mm ±SD (%)		Group B (RPF) Mean in mm ±SD (%)		P- value
	A <sub>1</sub> MTA	A <sub>2</sub> Biodentine	B <sub>1</sub> (MTA)	B <sub>2</sub> Biodentine	
3 months	0.06±0.02 (3.2%)	0.11±0.02 (5.5%)	0.07±0.021(3.19%)	0.10±0.01(5.52%)	0.001**
6 months	0.12±0.02 (6.36%)	0.18±0.04 (8.9%)	0.11±0.03(6.33%)	0.19±0.04(8.97%)	0.004*

\* The mean difference is significant at  $P \leq 0.05$  level.

\*\* is highly significant at the 0.001 level of significance ( $P \leq 0.001$ ).

**Table (4)** Comparison between the change in root dentin thickness and the percentage change in length at a different evaluation period within each group

Time \ Group	3 months		6 months		P-value
	1(MTA)	2(Biodentine)	1(MTA)	2(Biodentine)	
Group A	0.06±0.02 (3.2%)	0.11±0.02 (5.5%)	0.12±0.02 (6.36%)	0.18±0.04 (8.9%)	0.001**
Group B	0.07±0.021(3.19%)	0.10±0.01(5.52%)	0.11±0.03(6.33%)	0.19±0.04(8.97%)	0.001**

\* is significant at the 0.05 level of significances ( $P \leq 0.05$ ),

\*\* is highly significant at the 0.001 level of significance ( $P \leq 0.001$ )

### III. Change in apical diameter:

Table (5) shows the mean change in apical diameter and percentage of change in each group at each interval.

#### Group A1&B1 (MTA)

After 3 months, in group A<sub>1</sub>, the mean decrease in apical diameter was 0.07±0.03 mm representing 4.9% while in group B<sub>1</sub>, it was 0.068±0.028mm representing 4.18%. After 6 months, the mean decrease in apical diameter was 0.19±0.05 mm representing 14.1% in group A<sub>1</sub> and 0.18±0.04mm representing 13.398%.

After 3 months, the mean decrease in apical diameter was 0.09±0.03 mm representing 7.1%. After 6 months, the mean decrease in apical diameter was 0.22±0.07 mm representing 16.5% in

A<sub>2</sub> group and 0.21±0.06 mm representing 16.39% in B<sub>2</sub> group.

Statistical analysis showed that there was no statistically significant decrease in the mean apical diameter between the two groups through all follow up period.

Table (6) shows the mean change in apical diameter and percentage of change at different time periods within each group.

Regarding to the evaluation period at each group, there was significant decrease in mean apical diameter at the MTA group along the follow up periods. As well as, there was significant decrease in the mean apical diameter along the follow up periods at the Biodentine group.

**Table (5)** Comparison of changes in apical diameter and percentage of change in apical diameter in the two groups at each interval:

Time \ Group	Group A (Blood clot) Mean in mm ±SD (%)		Group B (RPF) Mean in mm ±SD (%)		P- value
	A <sub>1</sub> MTA	A <sub>2</sub> Biodentine	B <sub>1</sub> (MTA)	B <sub>2</sub> Biodentine	
3 months	0.07±0.03 (4.9%)	0.09±0.03 (7.1%)	0.068±0.028(4.18%)	0.09±0.02(7.09%)	0.208
6 months	0.19±0.05 (14.1%)	0.22±0.07 (16.5%)	0.18±0.04(13.398%)	0.21±0.06(16.39%)	0.396

\* The mean difference is significant at  $P \leq 0.05$  level.

\*\* is highly significant at the 0.001 level of significance ( $P \leq 0.001$ ).



**Table (6)** Comparison between change in apical diameter and percentage change in different evaluation period within each group

Time	3 months		6 months		P-value
	1(MTA)	2(Biodentine)	1(MTA)	2(Biodentine)	
Group A	0.07±0.03 (4.9%)	0.09±0.03 (7.1%)	0.19±0.05 (14.1%)	0.22±0.07 (16.5%)	0.001
Group B	0.068±0.028(4.18%)	0.09±0.02(7.09%)	0.18±0.04(13.398%)	0.21±0.06(16.39%)	0.001

\* is significant at the 0.05 level of significances ( $P \leq 0.05$ ),

\*\* is highly significant at the 0.001 level of significance ( $P \leq 0.001$ )

## DISCUSSION

Pulp necrosis of immature teeth caused by dental caries or trauma can stop the development of roots, resulting in roots with thin canal walls and open walls<sup>(13)</sup>. Management of immature necrotic teeth is a major challenge for the doctor. Thin and fragile blunderbuss canals in premature non vital teeth are extremely difficult to endodontic therapy, as it does not allow the use of many mechanical instrumentation, and it is difficult or impossible to close the open apex with traditional lateral condensation techniques or thermal plastic techniques<sup>(32)</sup>

These cases were treated with calcium hydroxide apexification<sup>(33)</sup>. However, they found that long-term use of calcium hydroxide had different drawbacks, multiple visits, and the chances of channel contamination between visits and increased dentine brittleness due to a proteolytic reaction and thus an increased risk of fracture. MTA apical pulg has been shown to be a good option for these cases. It has the advantage of a low number of visits and a high success rate, but the problem of weak thin dentinal walls persists.

The conventional apexification with long-term calcium hydroxide must be changed every 3 to 6 months for the hard tissue barrier to form at the apex. Modern treatments have used an artificial barrier from MTA. Both techniques are followed by conventional root canal filling, but do not increase wall fracture resistance, as the fragile thin blunderbuss canals are not reinforced<sup>(34)</sup>

Tissue regeneration rather than artificial replacement is an emerging and exciting field in health sciences<sup>(35)</sup>. The revascularization procedure is a biologically based alternative treatment that has been applied recently for, non-vital immature teeth<sup>(36)</sup>. Revascularization of non-vital, infected, and immature teeth has been approved to induce regeneration of preapical tissue to stimulate apexogenesis, and is documented as a new treatment for these teeth<sup>(37)</sup>.

Due to the importance of a bacteria-free environment, MTA was used in this study because this substance had excellent sealing capacity. It has been shown to prevent microbial leakage, biocompatible with the pulp tissue, and allow exceptional marginal adaptation<sup>(12,38)</sup>.

A mixture of antibiotics (ciprofloxacin, minocycline, and metronidazole) has been used to disinfect the affected necrotic root canals. Removal of bacteria from the canal, especially in the deeper layers of infected root, plays a key role in revascularization success. In agreement with other researchers, the critical step in regenerative therapy was the complete elimination of the root canal area with abundant irrigation, minimal instrumentation, and the placement of mixture of antibiotic baste effective in destroying common pathogens in the affected root canals, both in vivo and in vitro<sup>(39,40&41)</sup>.

Selection of the teeth was limited to the upper anterior necrotic teeth that are immature for patients, ranging in age from 9 to 12 years. Preoperative

and postoperative radiograph was standardized in dimensions using the Rinn XCP (Rinn XCP Alignment System) paralleling technique. Nagy et al., 2014<sup>(42)</sup> stated that this technique allows for the standardization of radiometric measurements for accurate quantitative assessment of treatment outcomes; root length and root thickness. Patients received a revascularization protocol treatment similar to the protocol described by Iwaya et al., 2001<sup>(43)</sup>, and a decision was made to use revascularization therapy rather than apexification to increase the length and thickness of root walls. Teeth with an apical foramen above 1.1 mm have been reported as suitable candidates for revascularization<sup>(44)</sup>.

In our results, patients were asymptomatic after the disinfection visit, and the clinical examination showed no signs of edema, erythema, or sinus tracts. Teeth were not sensitive to percussion and palpation, possibly due to the effectiveness of TAP against endodontic microorganisms, agrees with Nagy et al., 2014<sup>(42)</sup>.

We found that the results of the revascularized tooth response for the electric pulp test were not applicable, because patients did not give the same response when using another pulp test such as the hot or cold pulp tester. Petrino et al., 2010<sup>(45)</sup>, some patients were found to lack response to vitality testing, while Khetarpal et al., 2013<sup>(46)</sup> found a positive response and reported that revascularized teeth respond positively to cold testing. Law, 2013<sup>(47)</sup> noted that the absence of a pulp response does not necessarily indicate a lack of vitality.

Regarding to radiographic changes, there was complete healing of the radiolucent lesions in all the patients, this may be due to effective canal disinfection and appropriate coronal tight seal, this finding is in agreement with Moreno-Hidalgo et al., 2014<sup>(48)</sup>.

With regard to changes in root length, there was an increase in root length but without a statistically

significant difference between the all main groups and sub groups<sup>(9)</sup>.

In terms of changes in root dentine thickness, there was highly statistically significant increase in root dentin thickness at Biodentine groups (Blood clot & PRF) compared with the MTA groups, and this result may be due to the difference in the speed of chemical reaction during the setting of the material. Although both materials yield the same chemical compounds, this reaction may be faster in Biodentine, which has a faster setting time and a higher push-out bond strength at 24 hours<sup>(26&42)</sup> and this is agreement with Aldakak et al., 2016<sup>(49)</sup> and De Rossi et al., 2014<sup>(50)</sup> who reported that the only difference in the pulpal response to Biodentine and MTA was in the thickness of the mineralized tissue bridge, which was greater in case of Biodentine. For changes in apical diameter, there was a decrease in apical diameter in all groups without a statistically significant difference between the two groups during the entire follow-up period.

MTA, also because of its chemical, biological, and physical properties, its ability to release calcium ions, its setting in the presence of blood or water, and its high biocompatibility. For a good seal, a layer of 3-4 mm is recommended directly in contact with the blood clot, thus forming a first filling layer<sup>(16&23)</sup>. However, due to its gradual deterioration and poor sealing, MTA was gradually replaced by Biodentine, which has the ability to complete the formation of hard tissue for the tooth and has some advantages over MTA (similar characteristics of human dentin, no teeth discoloration).

Several researchers have attempted to explain the mechanism of revascularization. Some have reported that a small amount of vital pulp tissue containing dental stem cells (DPSCs) remains in the apical portion of the root canal. These dental pulp stem cells retain the ability to tissue regeneration and can proliferate into newly formed blood-clot matrix, differentiating into odontoblasts, and depositing third-degree or tubular dentin<sup>(51)</sup>.



Another explanation is the presence of stem cells in the periodontal ligament and bone marrow, when released using an over instrumentation, that can proliferate and grow at the apical region and within the root canal, depositing hard tissue both in the apical portion and on the sides of root walls<sup>(38&39)</sup>.

The more plausible explanation for the mechanism of revascularization is stem cells that residing in the apical papilla of undeveloped teeth. Because of its apical location, the papilla has collateral circulation that enables it to survive during the process of pulp necrosis. The step of inducing a hematopoietic column leads to a large accumulation of stem cells into the canal space. Furthermore, it contributes to the regeneration of pulpal tissues, and under the influence of epithelial cells surviving from epithelial heath of Hertwig's roots; it can differentiate into primary odontoblasts to continue root formation<sup>(27,40&41)</sup>.

## CONCLUSION

Although pulp revascularization is a newer treatment for regenerative endodontic procedures, it seems effective for immature teeth because it allows root formation in a relatively simple manner and enhance prognosis for treated teeth. In addition, placing Biodentine and MTA cement provided a good seal and favorable outcome. However, more studies are needed to assess their long-term effectiveness and new approaches.

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## إعادة تجدد الأوعية الدموية القمي للأسنان الدائمة المنخوره بطرق مختلفة

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### الملخص:

**الهدف:** لتقييم تأثير BIODENTINE و MTA على طول الجذر وسمك عاج الجذر والقطر القمي بعد إعادة تكوين الأوعية الدموية في الأسنان الدائمة غير الناضجة.

**المواد والأساليب:** أظهرت 20 سنًا أمامية علوية جذورًا غير ناضجة ذات قمة مفتوحة . تتراوح أعمارها بين 9 و 12 عامًا. تم إجراء علاج إعادة تكوين الأوعية الدموية . والذي بدأ بتطهير القنوات (الري هيبوكلووريت الصوديوم . متبوعًا بمعجون مضاد حيوي ثلاثي لمدة 2-3 أسابيع). بعد ذلك . تمت إزالة معجون المضاد الحيوي الثلاثي عن طريق الري بالمحلول الملحي . وتم تقسيم الأسنان إلى مجموعتين 10 كل مجموعة A: التنشيط باستخدام سقالة جلطة الدم فقط . والمجموعة B: التنشيط باستخدام الفيرين الغني بالصفائح الدموية والجلطة الدموية (PRF). تم تقسيم كل مجموعة إلى مجموعتين فرعيتين وفقًا للمادة الموضوعه فوق فتحة قناة الجذر وختم إكليلي من الزجاج الشارد. تمت المتابعة لمدة 6 أشهر. تم تحليل الصور الشعاعية المعيارية للشفاء حول الجذور وإغلاق قمي.

**النتائج:** كان هناك انخفاض كبير في متوسط القطر القمي على طول فترات المتابعة. لم تظهر فروق ذات دلالة إحصائية بين جميع المجموعات.

**الخلاصة:** أظهرت الأدلة السريرية والشعاعية أن إجراء إعادة تكوين الأوعية الدموية يمكن أن يكون علاجًا بديلًا للأسنان غير الناضجة. بالإضافة إلى ذلك . يوفر وضع الأسمنت BIODENTINE و MTA ختمًا جيدًا ونتائج مواتية.

**الكلمات المفتاحية:** بيودنتين . MTA . غير ناضج . أسنان دائمة . إعادة نوعية دموية

