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# Apical Closure in a Necrotic Immature tooth by Revascularization therapy using Platelet-Rich Fibrin: a Case Report

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### ARTICLE INFO

### ABSTRACT

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Keywords Pulp revascularization Apexification Dental pulp necrosis Platelet-Rich-Fibrin Immature tooth **Background and Aim:** Revascularization therapy by using Platelet-Rich Fibrin can be a suitable choice for revitalization of immature necrotic teeth as it improves root formation process, thickening of dentinal walls and apical closure.

**Case Presentation:** A 13-year-old girl with a history of trauma in tooth #21 and signs of pulp necrosis referred to the endodontics department of faculty of dentistry of Islamic Azad University. Intraoral radiograph showed immature root with open apex and periapical rarefaction. Access cavity was prepared and the root canal was irrigated with 20 ml of 1/25% sodium hypochlorite solution for 5 minutes and with normal saline. Equal proportions of Ciprofloxacin (tablet, 500mg), Metronidazole (tablet, 500mg) and Cefaclor (capsule, 500mg) were mixed and placed inside root canal for 4 weeks. Then, the canal was irrigated with 17% EDTA. 8 ml of patient's whole blood was centrifuged for preparation of PRF clot. PRF clot was placed inside the root canal up to CEJ level. MTA of 3mm thickness was placed directly over the PRF clot and tooth was temporarily restored. After 3 days, the tooth was double sealed with Glass Ionomer cement and composite resin. Clinical examinations at 1, 3 and 6 months after, revealed the resolution of periapical lesion, further root development and apical closure.

**Conclusion:**PRF clot is an ideal scaffold for regenerative endodontic therapy in necrotic immature teeth as it contains growth factors and can enhance cellular proliferation and differentiation of stem cells.

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### Introuduction

One of the major clinical challenges in root canal therapy, is the treatment of immature necrotic teeth. Trauma or pulp exposure due to dental caries can lead to pulp necrosis, infection and can impede root development. Following ceased root formation, dentinal walls remain thin and susceptible to fracture. Also, open apex is common in these teeth. The common treatment method for these teeth is formation of apical seal with MTA or Apexification procedure. In Apexification procedure calcium hydroxide paste is used to induce a calcified barrier. Despite the success of this treatment method, it has some disadvantages including long and repeated examination sessions (during 3 to 21 months), micro-leakage of the temporary restoration and cervical tooth fracture. <sup>1, 2</sup> Also, calcium hydroxide denatures dentinal collagen and decreases the root strength.<sup>2</sup>

MTA is recognized as a suitable material for apical seal formation in one or two sessions. But application of this material is difficult in teeth with widely open apex and blunderbuss dentinal walls. In these cases, condensing of MTA is difficult and protruding from the apex is probable. Also, in both mentioned methods, root length and thickness of dentinal walls remain unchanged and dental root remains weak and susceptible to fracture.<sup>1</sup>

In 2004 Banchs et al. performed a study on revascularization therapy which revolutionized endodontic treatments.<sup>2</sup> It seems that revascularization treatments are more suitable than previous treatments as they lead to root development, thickening of dentinal walls and apex closure. <sup>1</sup>In regenerative treatments, three factors play a major role: stem cells, growth factors and a suitable scaffold. In necrotic teeth, a suitable scaffold is necessary for release of growth factors and for forming a proper space for placement of stem cells. An empty root canal cannot provide a suitable support for proliferation and differentiation of stem cells. Recently, Torabinejad et al. have presented a case report on PRP (Platelet-Rich Plasma) as a scaffold in necrotic teeth treatment. <sup>3</sup>PRF (Platelet-Rich Fibrin) or the second generation of platelet aggregation was first introduced in 2001 by Choukroun et al. in France.<sup>4</sup> This material is an autogenic fibrin matrix which contains growth factor, platelet, leukocyte and cytokine.

PRF causes the remaining cells to proliferate. Even in extensive periapical lesions, a number of pulpal cells remain vital. After disinfection of root canal and decreased inflammation and due to the effect of Hertwig's epithelial root sheath on these cells, these remaining vital cells differentiate to odontoblast-like cells.<sup>5</sup>

### **Case Presentation**

A 13-year-old girl with a history of trauma in tooth # 21 referred to the endodontic department of dental faculty of Islamic Azad University. The patient was healthy and had a normal medical and dental history.

Intraoral examination revealed no abnormality or swelling in soft tissue. The mentioned tooth was not discolored and was not sensitive to cold and electric pulp tests. A mesial coronal fracture was evident due to last year trauma. In periodontal examination, probing depth was in the normal range. The radiographic examination showed periapical rarefaction, immature root with open apex. (Figure 1)

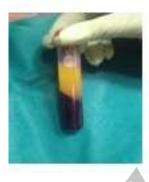


**Figure 1-Primary radiograph** 

Revascularization therapy with PRF was considered for this tooth and the treatment was initiated after receiving an informed consent from the patient's parents.

After Mepivacaine anesthetic injection, the tooth was isolated with rubber dam and access cavity was prepared. The root canal was irrigated with 20 ml of 1.25% sodium hypochlorite solution for 5 minutes and with normal saline and was dried with paper point.

As the root canal contained microorganisms that prevent revascularization, equal proportions of Ciprofloxacin (tablet, 500mg), Metronidazole (tablet, 500mg) and Cefaclor (capsule, 500mg) were mixed with normal saline to form a paste. Note that the sugar coated layers on tablets were removed. This antibiotic paste was placed in root canal for disinfection for 4 weeks. As the patient was non-symptomatic after this time period, the access cavity and root canal were irrigated with normal saline and 20 ml of 17% EDTA. For PRF isolation, 8 cc of venous blood was collected from CUBITAL vein in sterile tubes and was centrifuged for 12 minutes with speed of 2400 rpm. After centrifuge, three parts were formed inside each test tube: base layer containing erythrocytes, intermediate layer of PRF clot and surface layer of acellular plasma. (Figure 2)



# Figure 2- Contents of blood-containing test tube after centrifuge

PRF was divided to small pieces on a glass slab and was placed inside root canal with a hemostat and was condensed in apical direction with a hand plugger. Then, the canal orifice was filled with 3 mm thickness of MTA (PPH CERKAMD, Medical Company, Poland) to CEJ level. MTA was placed in direct contact with PRF and moist cotton ball and temporary dressing were placed on MTA. After 3 days the patient showed no signs of pain or swelling and after ensuring of MTA setting , the tooth was double sealed with self-curing Glass ionomer cement (GC Universal Restorative, GC Corporation, Japan) and com posite resin (3M ESPE, Universal Restorative, 3M ESPE dental products, USA). The tooth was non-symptomatic on 1, 3 and 6 months follow ups. Decreased periapical rarefaction was evident on radiograph in the first month (Figure 2) and it was minimized in the third month (Figure 3)



Figure 3 - the first month follow-up, periapical rarefaction compared to the primary radiographic image

and was totally resolved in the sixth month of follow up. Also, thickening of apical dentinal walls, increasing closure was seen during



Figure 4 - resolving periapical rarefaction and bone formation in the area

### Discussion

Revascularization therapy in immature teeth is a procedure that causes the root formation process to continue after disinfection of root canal and placement of new tissue. Therefore, infection control in this treatment method is of high importance. Hence, in this study 20 ml of 1.25% sodium hypochlorite was used for 5 minutes accompanied by normal saline. Also, application of 17% EDTA has been advised prior to PRF placement in the root canal. <sup>6</sup> EDTA causes release of growth factors from dentinal walls which leads to proliferation and differentiation of stem cells. <sup>7</sup>

In previous studies, an antibiotic mixture of Metronidazole, Ciprofloxacin and Minocycline was used but in the present study Cefaclor substituted Minocycline because Minocycline causes dental coronal discoloration but Cefaclor causes no discoloration which is in line with our study.<sup>15</sup>

After disinfection, a suitable scaffold is required for stem cell proliferation and diffusion of growth factors. An empty root canal cannot provide this suitable support for cell differentiation.<sup>8</sup> Thibodeau stated that revascularization with blood clot has shown satisfactory results in animal samples. Nevertheless, this method is accompanied by patient discomfort. Also bleeding control and placement of MTA on blood clot is difficult. Formation of blood clot is sometimes difficult and is not always possible. For example when calcium hydroxide is used between treatment sessions, it causes Coagulation necrosis. <sup>9</sup>

Collagen has also been used as a scaffold for revascularization therapy but this scaffold is inactive, contains no growth factors and does not have a role in stimulating cell proliferation. <sup>8</sup> PRP (Platelet-Rich Plasma) or the first generation of platelet aggregation is introduced as a suitable scaffold for revascularization. It contains growth factors and stimulates cell proliferation. Considering that PRP needs bovine thrombin or calcium sulfate for activation and bovine thrombin can cause unwanted reactions including hemorrhage, thrombosis and auto-immune reactions such as Systemic Lupus Erythematosus, it seems that PRF is a more suitable technique in revascularization therapy.

Nowadays, PRF (Platelet-Rich Fibrin) or the second generation of platelet aggregation is introduced as a suitable scaffold for revascularization. This material differs from PRP. PRF preparation is very simple in contrast to that of PRP and does not need any anti-coagulation material and therefor is an autogenic and natural substance. <sup>5</sup> This flexible scaffold has the ability to diffuse growth factors such as PDGF, TGF 1, VEGF and IGFS over a long period. Secretion of growth factors continues after this time period and gradually decreases. Diffusion of PDGF and TGF 1 continues for at least 7 to 28 days. <sup>8</sup> Also, presence of leukocytes and cytokines and few lymphocytes in PRF eases the infection and inflammation control. Angiogenesis is increased due to release of VEGF which plays an important role in revascularization.

PRF functional theory is stimulating stem cell proliferation and higher expression of Osteoprotegrin proteins and alkaline phosphatase. These proteins are usually identified as odontoblast differentiation markers. <sup>5</sup> MTA has been recognized as a suitable sealant and in this study was placed in direct contact with PRF for coronal seal. <sup>1</sup>

## Conclusion

PRF clot is an ideal scaffold for revascularization therapy in necrotic immature teeth as it contains growth factors and can enhance cellular proliferation, increase angiogenesis and it has anti-inflammatory effect. Also it is a suitable matrix for support of MTA and permanent coronal restoration and provides a proper dental seal.

### **Conflict of interests**

Authors report no conflict of interest related to this study.

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