

## **REGENERATIVE ENDODONTIC THERAPY: REPORT OF TWO CASES WITH DIVERSE CLINICAL APPROACHES AND RESULT**

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### **ABSTRACT**

Treating necrotic immature permanent teeth through endodontic procedures often poses clinical challenges. However, with careful case selection, regenerative treatments can yield positive results. Yet, there remains a lack of consensus regarding the ideal disinfection protocol or method to ensure consistent clinical outcomes. This article showcases two cases of regenerative treatment in necrotic immature teeth. Each case utilized mineral trioxide aggregate (MTA) and Biodentine™ as coronal barriers along with different irrigants, resulting in distinct clinical outcomes.

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### **INTRODUCTION**

Treating necrotic immature teeth in endodontics presents significant challenges. Achieving proper preparation and obturation of the apical portion is particularly difficult due to the thin, fragile dentinal walls and the complex blunderbuss anatomy<sup>1</sup>. In the past, the preferred treatment for such cases was multiple-session apexification using calcium hydroxide<sup>2</sup>. Subsequently, a one-step apexification method was proposed, involving the induction of artificial barriers using materials like MTA. This approach significantly reduced the frequency of sessions and shortened the duration of treatment<sup>3</sup>. Nonetheless, both of these techniques have a significant drawback: they do not support continued root development, leading to the formation of a fragile root<sup>4</sup>. A novel approach recently proposed involves creating an environment conducive to root maturation. This method includes disinfecting the root canal system and using antibiotic paste as an intracanal medicament. Revascularization, as a biological alternative, offers a different approach for treating immature necrotic teeth.

Unlike conventional apexification and artificial formation of an apical barrier, revascularization facilitates root maturation<sup>5,6</sup>

In cases of necrotic, open-apex teeth, it's possible that vital apical pulp tissue or Hertwig's epithelial root sheath may still be present. If these tissues exist, they have the potential to proliferate and contribute to root development, particularly when the canal has been thoroughly disinfected, and the inflammatory process has been reversed.<sup>7</sup>

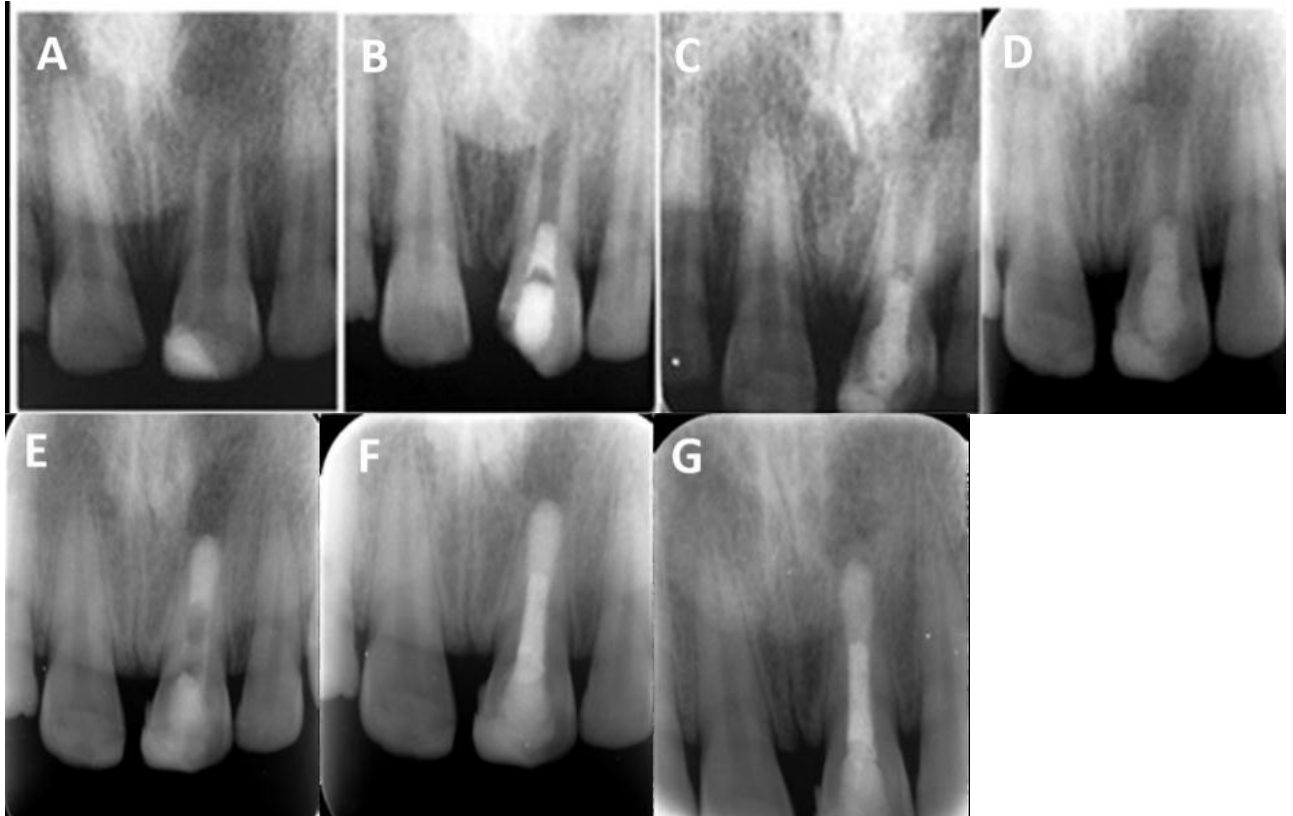
Modern era of regenerative endodontics was started by a case report by Banchs and Trope<sup>8</sup> in 2004.

The proposed regenerative treatment generally starts with chemical disinfection of the root canals. Different concentrations of sodium hypochlorite (NaOCl) including 6%<sup>9,10</sup>, 5.25%<sup>5,8,11</sup>, 2.5%<sup>12,13</sup>, and 1.25%<sup>14</sup> and different concentrations of chlorhexidine (CHX) including 2%<sup>9</sup> and 0.12%<sup>11</sup> have been successfully used for this purpose. The procedure continues by application of triple antibiotic paste as an intracanal medicament, which is composed of ciprofloxacin, metronidazole and minocycline, as suggested by Hoshino et al<sup>15</sup>. If there are no clinical signs or symptoms of periradicular diseases, the treatment progresses by removing the paste and inducing bleeding into the canals by irritating the periapical tissues. Once a blood clot forms, the canal's orifice is sealed with MTA, a biocompatible sealing material placed over the clot. Finally, the crown is permanently restored to complete the procedure. It's important to note that revascularization has practical limitations. There is currently no consensus on methods to consistently achieve predictable clinical outcomes or establish optimal disinfection protocols. Various clinical protocols have been employed, utilizing different irrigants, medicaments, procedures, and follow-up schedules. Additionally, criteria for reliably predicting successful revascularization are still lacking. Selecting suitable non-vital teeth with residual vital apical cells, considered crucial for a successful regenerative procedure, poses a challenge due to this uncertainty. The article delineates two distinct revascularization treatment protocols for necrotic immature teeth, each resulting in varied clinical outcomes.

## **CASE REPORT**

CASE ONE: A healthy 16-year-old male patient was referred to the Department of Endodontics at Rama Dental College, Hospital, and Research Centre. The patient reported occasional pus discharge from a gumboil in the anterior region of the upper jaw. He recalled experiencing an impact trauma and crown fracture of the left maxillary incisor eight years

prior. The patient reported a previous history of an impact trauma and crown fracture of the left maxillary incisor eight years ago. Upon clinical examination, crown fracture and a composite filling on tooth #21 were observed. Tooth mobility was within the normal limits. All the teeth in the maxillary anterior region were responsive to cold test, using Endo-Frost cold spray (Roeko; Coltene Whaledent, Langenau, Germany) except for tooth #21. There was no traceable sinus tract at the time. Radiography revealed immature root of tooth #21 with a radiolucent periapical lesion (Fig. 1A). The diagnosis of necrotic pulp with asymptomatic apical periodontitis was made. Informed consent was obtained. After local infiltration anesthesia with 1.8 mL of 2% lidocaine with 1:80,000 epinephrine (Daroupakhsh, Tehran, Iran), rubber dam isolation, and access cavity preparation, the working length was determined by placing a large file in the canal and taking a periapical radiograph. The root canal system was irrigated with 20 mL of 5.25% NaOCl followed by 20 mL of 0.2% CHX<sup>8,16</sup>. Triple antibiotic paste (ciprofloxacin, metronidazole, doxycycline) was used as intracanal medicament for three weeks. In the next visit, the patient was asymptomatic. Local infiltration anesthesia was performed with 3% plain mepivacaine, without vasoconstrictor to facilitate bleeding as suggested by Petrino et al<sup>11</sup>. The antibiotic intracanal medicament was gently removed and flushed out of the canal with copious irrigation with 5.25% NaOCl. After drying the canal, bleeding was induced inside the canal with a sterile #50 hand file (MANI Inc., Utsunomiya, Japan), which was inserted one millimeter beyond the apical foramen and the coronal part of the canal was sealed with ProRoot MTA (Dentsply Tulsa Dental, Tulsa, OK, USA) over the blood clot. A moist cotton pellet was placed over the MTA in the access cavity, and the tooth was temporarily restored with Coltosol (Asia Chemi Teb Co., Tehran, Iran) (Fig. 1B).



**Fig. 1.** (A) Preoperative periapical radiograph of tooth #21. Note the immaturity of root. (B) After placing MTA on blood clot and temporary filling. (C) After completion of regenerative endodontic procedure and permanent coronal restoration. (D) Six-month follow-up. Note the periapical radiolucent lesion. (E and F) After placing MTA plug as apical barrier and obturation of coronal part of the root. (G) One-year follow-up. The healing of periapical lesion was observed.

A week later, the patient returned for a follow-up appointment to confirm the setting of MTA. Subsequently, permanent restoration of the tooth was carried out using composite resin (Dentsply International, Milford, DE, USA). Three months later, at the initial follow-up appointment, no signs or symptoms were noted. However, during the six-month recall, the patient reported a recurrence of pus discharge. Radiographs did not reveal any evidence of thickening of the canal walls or continuation of root development. Apical rarefaction was observed (Fig. 1D). According to the American Association of Endodontics (AAE) guidelines<sup>17</sup>, the regenerative treatment was considered not successful. Therefore, apical closure with MTA plug was scheduled. After preparing the access cavity, the coronal MTA was removed with a diamond-coated straight tip (E32D, NSK, Tokyo, Japan) attached to an ultrasonic scaler (Varios 970, NSK, Tokyo, Japan) under copious irrigation. A five-millimeter thick ProRoot MTA plug was placed in the apical part of the canal (Fig. 1E). After one week, the MTA setting was ensured and the coronal part was filled with gutta-percha (Meta Bio-med

Co., LTD, Seoul, Korea) and AH-26 sealer (Dentsply, De Trey, Konstanz, Germany). The crown was restored with composite resin (Dentsply International, Milford, DE, USA) (Fig. 1F). The patient underwent regular recall appointments every three months for radiographic examination and assessment of clinical signs and symptoms. Throughout the follow-up sessions and at the one-year mark, the patient remained asymptomatic with no evident signs of disease. At the one-year follow-up, the radiolucent lesion had resolved (Fig. 1G).

**CASE TWO:** A healthy 17-year-old female patient was referred to the Department of Endodontics at Rama Dental College, Hospital and Research Centre. She reported slight swelling and pus discharge adjacent to one of her lower teeth. Upon clinical examination, an opening was noted on the occlusal surface of the mandibular right second premolar (tooth #45), which appeared to result in pulpal exposure (Fig. 2).



**Fig. 2.** Cavity on the occlusal surface of tooth #45 and a sinus tract in the lingual side of the tooth.



**Fig. 3.** Panoramic radiograph. Note the immaturity of tooth #45.

The tooth did not exhibit responsiveness to a cold test using Endo-Frost cold spray. Additionally, a swelling and sinus tract were observed on the lingual side of the tooth, although the sinus tract was not discernible. Radiographically, tooth #45 displayed a

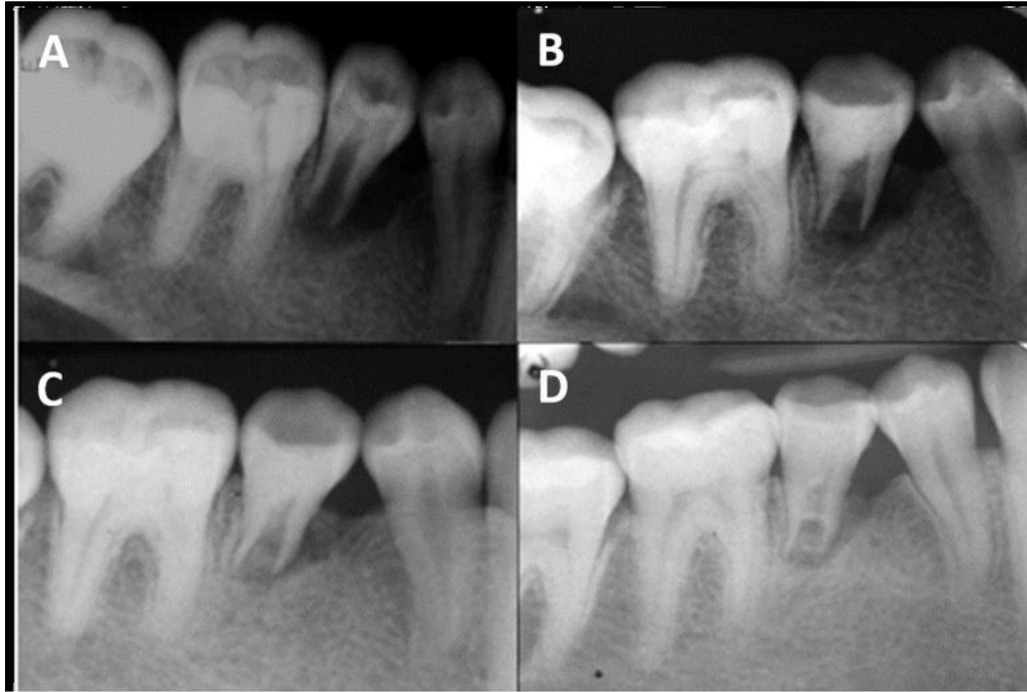
blunderbuss short root with thin dentinal walls and a radiolucency surrounding the root (Figs. 3 and 4A).

During the initial appointment, informed consent was obtained from the patient. Following this, local anesthesia was administered using a solution containing 3% plain mepivacaine, and rubber dam isolation was applied. Subsequently, the access cavity was prepared. The working length was established by placing a large file into the canal and taking a periapical radiograph for verification. Following this, the root canal system was thoroughly and gently irrigated with 20 mL of 1.5% sodium hypochlorite (NaOCl), followed by a rinse with 20 mL of normal saline<sup>14</sup>. Triple antibiotic paste (ciprofloxacin, metronidazole, doxycycline) was used as intracanal medicament for three weeks (Fig. 5). At the subsequent visit, the patient remained asymptomatic, and the sinus tract had resolved. Local infiltration anesthesia was administered using 3% plain mepivacaine. The antibiotic intracanal medicament was carefully removed from the canal through irrigation with 20 mL of normal saline followed by 20 mL of 17% EDTA solution. Subsequently, the canal was dried, and bleeding was induced inside the canal using an overextended #70 hand file (MANI Inc., Utsunomiya, Japan). Finally, the coronal part of the canal was sealed Biodentine™ (Septodont, St. Maur-des-Fossés, France). One week later, the tooth was permanently restored with composite resin (Dentsply International, Milford, DE, USA) (Figs. 4B and 6). The patient underwent recall appointments every three months, during which no clinical signs or symptoms were observed. At the six-month follow-up, radiographs did not reveal any thickening of the canal walls or continuation of root development. However, notable intracanal calcification was observed (Fig. 4C). The patient remained asymptomatic with no signs or symptoms observed at the one-year follow-up, mirroring the clinical findings from previous visits. Radiographically, diffuse intracanal calcification was evident around the coronal Biodentine™, with radiopaque calcified bridges noted in the middle and apical portions of the canal (Fig. 4D). The tooth did not respond to cold testing.

## **DISCUSSION**

In this case report, we examined two immature necrotic teeth presenting with periapical radiolucencies. Both patients were of the same age. In the first case, pulp necrosis was attributed to a traumatic incident. Conversely, in the second case, pulp exposure was suspected to be caused by caries or dens evaginatus, resulting in necrosis before tooth

maturation. Dens evaginatus can manifest in any tooth but is frequently observed in premolars, particularly mandibular premolars.



**Fig. 4.** (A) Blunderbuss short root of tooth #45 with thin dentinal walls and a radiolucency embracing the root. (B) After regenerative endodontic treatment and permanent coronal restoration. (C) Six-month follow-up. (D)

Dens evaginatus stands as one of the most common tooth anomalies, often resulting in pulp exposure and subsequent devitalization at an early age due to attrition during regular tooth function.<sup>8,11,18</sup> In cases where necrosis occurs, they typically remain asymptomatic, with the potential development of periapical lesions and the formation of sinus tracts.

Pulp revascularization was deemed the treatment of choice to preserve the teeth and facilitate root development. The principal objective of regenerative endodontic procedures aligns with the healing of apical periodontitis, as outlined in the revised AAE guidelines (July 2013). According to the guidelines, the secondary goal is to enhance root wall thickness and/or root length, while the tertiary goal is to restore a positive response to pulp testing. Although both the secondary and tertiary goals are desirable, they may not be deemed essential for determining clinical success<sup>17</sup>. According to the guidelines, the first case did not meet the essential criteria for successful treatment, leading to the implementation of an alternative treatment plan.

Root canal disinfection in regenerative endodontic treatment remains challenging and subject to debate. One crucial aspect is selecting the most effective antimicrobial agent to ensure thorough cleaning of the root canal system. Additionally, in teeth with an immature apex, the root canal tends to be larger in size, facilitating easier permeation of antimicrobial agents into the root canal system and towards the periapical region.

Indeed, the primary concern lies in preventing relatively toxic antimicrobial agents from reaching the periapical tissues, which may harbor essential stem cells and vasculature crucial for the regeneration process. Efforts have been directed towards identifying antimicrobial agents with minimal toxic effects, considering both their chemical composition and concentration. The utilization of EndoVac has been highlighted as an effective strategy to mitigate periapical extrusion of irrigants like sodium hypochlorite (NaOCl)<sup>19</sup>. The different concentrations of NaOCl including 6%<sup>9,10</sup>, 5.25%<sup>5,8,11</sup>, 2.5%<sup>12,13</sup> and 1.25%<sup>14</sup> and different concentrations of CHX (0.12% to 2%)<sup>9,15</sup> have been successfully used for this purpose. In our two cases, different irrigation protocols were used. In the first case, the tooth was irrigated with 5.25% NaOCl followed by 0.2% CHX<sup>8,16</sup>.



**Fig. 5.** End of first appointment. Note the non- traceable sinus tract in the lingual side of tooth #45.



**Fig. 6.** After permanent restoraion

For removing the triple antibiotic paste in the second session, irrigation was performed using 5.25% NaOCl.

In the second case, the root canal system was irrigated with 1.5% sodium hypochlorite (NaOCl), followed by a rinse with normal saline<sup>14</sup>. During the subsequent visit, the antibiotic dressing was removed from the canal using normal saline. Following this, 17% EDTA was employed as the final irrigant, as EDTA is known to provide dentin-derived growth factors,



which can be beneficial for the regenerative process<sup>20,21</sup>. Research indicates that half- or full-strength concentrations of sodium hypochlorite (NaOCl), at 3% and 6%, respectively, have been shown to prevent stem cell attachment to dentin surfaces and are toxic to stem cells of the apical papilla (SCAP)<sup>20,21</sup>. Additionally, it has been demonstrated that 2% chlorhexidine (CHX) was the most toxic irrigant for SCAP. It's worth noting that in some successful revascularization cases reported in literature, full-strength NaOCl has been utilized for irrigation, at least during the initial appointment<sup>8,11</sup>. Previous studies showed that the use of 17% EDTA significantly increased attachment of newly formed mineralized tissues to dentinal canal walls<sup>21</sup>. Indeed, growth factors are released from the root canal dentinal walls following EDTA irrigation. In the second case, the decision to eliminate chlorhexidine (CHX) from the rinsing protocol and instead utilize 17% EDTA, along with using sodium hypochlorite (NaOCl) in a lower concentration, may have positively influenced the treatment outcome.

In the second case, Biodentine™ was utilized as the coronal barrier instead of MTA. Biodentine™ is a bioactive tooth-colored cement based on calcium silicate. Previous research has demonstrated the bioactivity of Biodentine™, as it promotes pulp cell proliferation and biomineralization. Therefore, Biodentine™ has been recommended as a suitable material for facilitating dentin-pulp complex regeneration in clinical practice<sup>1,23</sup>. Only one previous study has utilized Biodentine™ in the revascularization process and reported the resolution of associated periapical pathology. This study involved a mandibular incisor in a 15-year-old patient<sup>1</sup>. In the study mentioned, the tooth was irrigated with 6% sodium hypochlorite (NaOCl), and triple antibiotic paste was applied as the intracanal medicament without instrumentation. Subsequently, Biodentine™ was used to achieve a coronal seal. The tooth was then permanently restored with bonded resin. Remarkably, the periapical lesion resolved after 18 months. The authors concluded that Biodentine™ was effective in maintaining the vitality of dental pulp stem cells and fostering an environment conducive to pulp revascularization, leading to the completion of root maturation<sup>1</sup>. The success of revascularization/revitalization therapy depends on efficient disinfection of the root canal system<sup>24</sup>. If infection persists within the root canal, both regeneration and repair processes may be impeded in the pulp-periapical tissue complex<sup>24</sup>. Additionally, there may be a correlation between the time of trauma and the quality of root development; prolonged duration of pulp necrosis could lead to poorer root development following regenerative

treatments. This association has been highlighted in previous studies, which have reported reduced or absent root development and subsequent procedure failure<sup>11,25</sup>.

Lenzi and Trope<sup>25</sup> explored the notion that chronic infection may eradicate cells capable of pulp regeneration. However, they suggested that this might not be the sole reason, given the successful outcomes observed in regenerative endodontic treatments involving long-standing apical periodontitis cases. Another potential explanation they proposed is the maturation of bacterial biofilm, making it more challenging to eradicate using conventional protocols. This could possibly serve as one of the underlying factors contributing to the unfavorable outcome observed in our first case.

In the second case, intra-canal calcification was observed without an increase in root length or thickness of dentinal walls. However, the patient remained asymptomatic. According to the AAE guidelines, the essential requirement for clinical success is the healing of apical periodontitis<sup>17</sup>. Additionally, deposition of a cementum-like tissue on the root canal dentinal walls following regenerative endodontic treatment was documented in an animal study. This irregular tissue was assumed to contribute to root development. Furthermore, the authors reported the formation of cementum bridges in the root canal system, potentially attributed to the hard-tissue induction potential of MTA<sup>26</sup>

Fouad and Nosrat<sup>27</sup> proposed the necessity for clinicians and the research community to establish a consensus regarding the clinically acceptable outcome. They suggested redefining clinical success to entail the occurrence of calcification without any signs or symptoms and complete resolution of infection. While the formation of intracanal mineralized tissue and pulp canal obliteration may not align perfectly with the concept of regeneration, they advocated for a pragmatic approach, acknowledging and accepting some imperfections in the pursuit of progress in this field.

## **CONCLUSION**

The ideal outcome of regenerative endodontic treatment is a subject of considerable debate. There appears to be a discrepancy between the anticipated histological outcomes and the actual events occurring within the root canal system in many cases. However, since numerous teeth achieve acceptable clinical outcomes, such as being infection-free, asymptomatic, and functionally sound, it seems reasonable to reconsider our perspectives in this field. Expanding the definition of success in endodontic regenerative treatment to include these clinical outcomes would be a logical step forward.

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