Revascularization of Dental Pulp in Human Necrotic Permanent Teeth with Immature Apex: three case reports

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Abstract: Introduction: Treatment of non vital infected immature teeth presents challenge for endodontic outcome. Revascularization of immature necrotic teeth is a reliable treatment alternative to conventional apexogenesis or apexification. The purpose of this study was to examine the effect of a pulpal revascularization procedure for immature necrotic teeth with apical periodontitis. Methods: Three patients, each with an immature permanent central incisor tooth with chronic or acute apical periodontitis, were recruited. A triantibiotic mix (ciprofloxacin, metronidazole, and minocycline) was used to disinfect the pulp for 2 weeks. Then a blood clot was created in the canal, over which grey mineral trioxide aggregate was placed. Patients were recalled periodically. Results: The treated teeth (n = 3) were found to exhibit complete root development, with a positive response to pulp testing. Conclusions: Revascularization could be effective for managing immature permanent teeth with apical periodontitis with appropriate case selection.

Keywords: revascularization, pulp necrosis, immature tooth.

1. Introduction:
Non vital infected immature permanent teeth present multiple challenges for successful endodontic treatment. The mechanical debridement with standard root canal instrument and NaOCl irrigant has been proven ineffective to achieve proper cleaning and disinfect the overall dentin wall, particularly in immature apex\textsuperscript{1}. The hermetic apical seal is very difficult to obtain without overfilling as a result of eliminating the apical constriction. Furthermore, the presence of thin apical root thickness provides a significant risk to root fracture\textsuperscript{2}. The traditional management of such cases is apexification technique using Calcium hydroxide\textsuperscript{3}, or Mineral Trioxide Aggregate (MTA)\textsuperscript{4-7}. This technique is successful in inducing apical closure. However, there is no expectation of root lengthening. The innovation in endodontic field would be the regeneration of a functional pulp-dentin complex as an alternative technology to replace the traditional apexification, in attempting to stimulate further root development and thickening of dentinal walls in a non vital immature tooth\textsuperscript{3-9}. Several studies have recently focused on the potential for revascularization of infected root canals if their environment has been improved with adequate disinfection. Windley \textit{et al.},\textsuperscript{10} determined the effectiveness of antibiotic paste in the disinfection of immature dog teeth with apical periodontitis. They suggested that predictable revascularization of necrotic immature teeth would be expected if three challenges can be met including canal disinfection, placement of scaffold matrix to permit tissue ingrowth and bacterial tight seal coronally.

Aim of the study
The purpose of this study was to examine the effect of a pulpal revascularization procedure for immature necrotic teeth with apical periodontitis.

2. Methods:
Three cases were selected from patients attending Pediatric Dentistry clinic of the dental School of King Abdul Aziz University. The teeth inclusion criteria would be: 1) tooth associated with necrotic pulp, as a result of trauma or carious pulp exposure, with signs and/or symptoms of periapical pathology. 2) The tooth should have an immature apex, either tubular or blunderbuss. The teeth will be evaluated clinically and radiographically to determine the preoperative status, regarding pulp sensitivity test percussion, and palpation sensitivity, tooth mobility, pocket depth, presence or absence of apical periodontitis, presence or absence of sinus tract.
At each follow up recall period, the patient will be assessed clinically as well as radiographically.

The treatment was considered successful according to the following criteria; clinical criteria including lack of signs and symptoms.

The radiographic criteria including: (1) evidence of periapical healing (if periapical lesion was present) (2) radiographic evidence of increased root length, and (3) radiographic evidence of increased root canal wall thickness.

3. Case Reports

Case 1

A 9-year-old boy was referred to the pedodontic department, Faculty of Dentistry, King Abdulaziz University, for evaluation and treatment of his maxillary central incisor. His medical history was unremarkable. His dental history revealed that the patient had traumatic injury 3 months ago affected his maxillary central incisor. Clinical examination revealed that the tooth #21 had broken incisal edge, was sensitive to percussion and not responsive to electric pulp test. Periodontal probing depths were within normal limits (< 3 mm). Radiographic examination revealed an immature tooth with periapical radiolucency. Based on the results of the clinical and radiographic examinations, a diagnosis of tooth #21 was made of pulp necrosis with symptomatic apical periodontitis.

Under local anesthesia, the tooth was isolated with a rubber dam and the access cavity was prepared. The root canal was irrigated with 10 mL of 2.5% sodium hypochlorite for 2 min, and then dried with sterile paper points. and filled with triple antibiotic paste in a 1:1:1 ratio of a creamy paste mixture of metronidazole (Samil Pharm, Seoul, Korea), ciprofloxacin (Sinil Pharm, Seoul, Korea) and minocycline (Aurobindo Pharma, USA Inc) in sterile saline was applied using a lentulo-spiral and tapped down into the canal with the blunt end of sterile paper points. The tooth was temporarily restored using Caviton (GC, Aichi, Japan).

The patient was asymptomatic 2 weeks later. The Caviton was removed under rubber dam isolation. The mixture of antibiotics was completely removed with 2.5% sodium hypochlorite and sterile saline. Under local anesthesia without vasoconstrictor the apical tissue was stimulated with a No. 20 K-file to induce bleeding. Control bleeding was performed at the level just below CEJ. A blood clot formed in the canal about 15 min after stimulation. Mineral trioxide aggregate (MTA) (Dentsply Tulsa Dental, Johnson City, TN, USA) was applied over the blood clot, followed by a moist cotton pellet (Figure 1b). Caviton was placed temporarily over the cotton pellet.

One week later, Caviton and the cotton pellets were removed and the coronal accesses were finally sealed with composite resin (Z250; 3M ESPE,St Paul, MN, USA). The patient instructed to recall 2 weeks and then every 6 months for postoperative follow up. At all follow-up evaluation, the patient was asymptomatic. The treated tooth was respond within normal limit to percussion and palpation versus the adjacent lateral incisors At 18-month follow-up The radiographs demonstrated evidence of periradicular bone healing and significant root development with maturation of the dentine as compared with the preoperative radiographs(Fig 1).

![Pre-operative](image1.png) ![2 weeks](image2.png) ![12 months](image3.png) ![18 months](image4.png)

**Figure 1:** (Case1) (A) Preoperative radiophotographs of showed tooth # 11 having open apex with periapical radiolucency. (B) Immediate postoperative radiophotograph.(C) Radiophotographs after 12 months showed disappearance of periapical radiolucency related to tooth. (D) At 18-month follow-up, further narrowing of root canal in the apical third and thickening of the lateral walls were evident; normal bony architecture at the periradicular region could be seen.
Case 2:

A 11-year-old boy was referred to the Pedodontic department, Faculty of Dentistry, King Abdulaziz University, for evaluation for evaluation of tooth # 11. The patient had previous dental trauma 2 years ago. On the clinical examination, the patient was asymptomatic, and the tooth appeared with uncomplicated crown fracture. There was no response to either cold test or electric pulp tester. No tenderness to percussion or palpation was noticed. Periodontal probing and mobility were within normal limit. Upon radiographic examination, the tooth had an open apex with periapical radiolucency. Under local anesthesia, the tooth was isolated with a rubber dam and the access cavity was prepared. The root canal was irrigated with 10 mL of 2.5% sodium hypochlorite for 2 min, and then dried with sterile paper points. The root canal was filled with triple antibiotic paste in a 1:1:1 ratio of a creamy paste mixture of metronidazole (Samil Pharm, Seoul, Korea), ciprofloxacin (Sinil Pharm, Seoul, Korea) and minocycline (Aurobindo Pharma, USA Inc) in sterile saline was applied using a lentulo-spiral and tapped down into the canal with the blunt end of sterile paper points. The tooth was temporarily restored using Caviton (GC, Aichi, Japan). Two weeks later the tooth was permanently restored with composite resin and the patient was scheduled for recall and advice to call if any complain. At the follow up periods 2 weeks and every 6 month, the tooth was asymptomatic. The sensitivity test was still inconclusive. The radiograph at the 6-month showed complete resolution of radiolucency and evidence of radiopaque area at the apex and adjacent to the apical level of MTA was obvious. After 18- follow-up, The radiographs demonstrated evidence of periapical bone healing and significant root development with maturation of the dentine compared with the preoperative radiographs (Fig. 2).

Figure 2: (Case 2) (A) Preoperative radiograph showed tooth # 11 with open apex with periapical radiolucency. (B) 2 weeks postoperative radiograph. (C) Follow-up radiograph at 12 months showed reduction of the periapical lesion and narrowing of the Apex. (D) Follow-up radiograph at 18 months showed normal periradicular architecture and significant root development with maturation of the dentine.

Case 3:

A 8-year-old girl was referred to endodontic clinic KAUFU for root canal treatment of tooth # 21. A review of her dental history revealed previous trauma 2 years ago affecting the complaint tooth. No previous endodontic treatment was done. No contributories were considered related to her medical status. Upon clinical examination, intraoral sinus tract on the gingival region of tooth # 21 was present. The electric pulp test and cold tests produced no response. The periodontal evaluation revealed no tenderness to neither percussion nor palpation tests. The tooth was within normal physiological mobility. A periapical radiograph was taken with gutta-percha point to trace the sinus tract (Fig 1 a). The sinus tract was found to be related to the periradicular radiolucency at the apex of tooth # 21. Based on clinical and radiographic examination, the tooth # 21 was diagnosed as necrotic pulp with chronic apical abscess. Four weeks after the anti-bacterial medication was placed on the tooth # 21 the patient was asymptomatic and without swelling or sinus tract. Under local anaesthesia and rubber dam isolation, the tooth was treated using the same clinical protocol used for the previous case. Sodium hypochlorite irrigation was used for removal of the anti-bacterial paste followed by stimulation of hemorrhage, clot formation and MTA placement. The tooth was then temporized and the patient was rescheduled for the final composite restoration which
was placed 2 weeks later without incident or change in symptoms. During the 18-month follow-up period the patient remained asymptomatic. No tenderness to percussion or palpation was noted and the periodontal examination revealed no pocket depths over 3 mm and normal physiological mobility. The radiographs demonstrated evidence of periradicular bone healing and significant root development with maturation of the dentine as compared with the preoperative radiographs (Fig 3).

Figure 3: (Case 3) (A) Preoperative radiophotograph showed tooth # 21 with open apex and periapical radiolucency. (B) 2 weeks postoperative radiophotograph. (C) Follow-up radiograph at 12 months showed decrease in the size of the periapical lesion and elongation of the root was evident. (D) Follow-up radiograph at 18 months showed disappearance of apical radiolucency and significant root development with maturation of the dentine.

4. Discussion
Pulp necrosis of an immature tooth as a result of caries or trauma could arrest further development of the root, leaving the tooth with thin root canal walls and open apices. Management of immature non vital teeth poses a great challenge to the clinician. A major challenge faced by most the endodontists is the treatment of thin, fragile blunderbuss canals in non vital teeth.

Endodontic treatment of such a tooth is difficult because the thin walls do not allow much mechanical instrumentation and the open apex is difficult or impossible to seal with conventional methods of lateral condensation or thermoplasticized techniques. The traditional treatment for these teeth is long-term calcium hydroxide application to induce apexification (an apical hard tissue barrier). More recent treatments have used an artificial barrier of mineral trioxide aggregate (MTA). Both of these techniques are followed by a traditional root filling, but they do not increase the fracture resistance of the walls as strengthening or reinforcing of the thin fragile blunderbuss canals is not achieved. Root-wall–strengthening methods with composite resin have been advocated but they may limit the possibility of root canal retreatment if the need arises in the future.

Regeneration of tissues rather than replacement with artificial substitutes is an emerging and exciting field in the health sciences. It is an alternative biologically based treatment which has been introduced for immature teeth with necrotic pulp. Procedures that preserve the remaining dental pulp stem cells and mesenchymal stem cells of the apical papilla can result in intracanal revascularization and continued root development. The revascularization of these teeth is based on the concept that vital stem cells in the apical papilla can survive pulpal necrosis, even in the presence of periapical infection, because the open apex provides good communication to the periapical tissues. Revascularization of infected, nonvital, immature teeth has been documented to stimulate regeneration of apical tissues and to induce apexogenesis and is emerging as a new treatment modality for such teeth.

The present case reports evaluated the prognosis of revascularization in necrotic immature teeth, with the aim of providing reliable evidences for the revascularization technique.

In the present study combinations of topical antibiotics used to disinfect necrotic, infected root canals as eradication of bacteria from the pulp canal plays a key role in successful revascularization, because the process will halt in the presence of infection. Sato et al., determined that this drug combination was effective for eliminating bacteria in the deep layers of root dentin. This in agreement with other researchers who stated that, a critical step in regenerative therapy is complete disinfection of the root canal space using copious irrigation, minimal instrumentation and placement of antibiotic pastes in
the presence of a suitable scaffold 23-27. Studies have shown that local application of antibiotics (mixture of ciprofloxacin, metronidazole, and minocycline) is effective in killing common endodontic pathogens in infected root canals, both in vitro and in vivo 28,29.

After disinfection step bleeding was induced into the root canals and blood clot was allowed to be formed to act as scaffold for tissue ingrowth as empty tube will not support the ingrowth of tissues from the periapical region. Thibodeau et al., 30 showed that roots containing a blood clot had better treatment outcomes in a dog model than those that did not have a blood clot in the apical part of the canal. Another studies stated that, besides acting as a scaffold, the blood clot might also contain growth and differentiation factors that might be important for successful revascularization of the empty pulp canal 31,32.

Given the importance of a bacteria-free environment, a coronal seal with MTA was used in this present study, which material has been shown to possess an excellent sealing ability 33,34.

All successful cases presented here showed an increased root length and narrowing of the canal space. However, it remains uncertain whether the thickened canal wall was actually made up of dentin. This can be explained by that the blood clot acted as a matrix onto which the vital cells from peri-apex could get seeded and reestablish the pulp vascularity. Some studies tried to describe various other mechanisms of vascularization. They stated that vital mature (adult) pulp cells remain at the apical end of the root canal and cause pulp revitalization; and, revascularization of pulp occurs because of multi-potent dental pulp stem cells, stem cells in the periodontal ligament, or stem cells from the bone marrow released by over-instrumentation. The most plausible mechanism of the root development is the isolation of stem cells from SCAP. It has been hypothesized that SCAP can be the source or primary odontoblasts forming the root dentin 35.

Another explanation stated that traces of pulpal tissue might survive apically, even in the presence of a large periapical lesion. In the case of an immature necrotic tooth with apical periodontitis, the stem cells from the apical papilla (SCAPs) might survive the infection and are capable of forming odontoblast-like cells, producing dentin in vivo. After root canal disinfection, under the influence of the surviving epithelial cells of the Hertwig’s root sheath, SCAPs can differentiate into primary odontoblasts to continue root formation 36,37.

The resultant increase in the root length and thickness of the canal wall would strengthen the tooth against fracture. With a fully formed apex, the prognosis for any root canal treatment that might become necessary (as a result of necrosis of the new “pulpal” tissue) in the future would be much better than any attempt to obturate an open apex 38.

Conclusion

In conclusion, revascularization induced maturation, where indicated, can provide several advantages over conventional apexification procedures. A detailed histo-pathological study is necessary to demonstrate the actual contents of pulp space after revascularization procedures.

Acknowledgement

This project was funded by the Dean of Scientific Research (DSR), King Abdulaziz University, Jeddah, under grant no.(105/254/1432). The authors, therefore, acknowledge with thanks DSR technical and Financial support.

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7/21/2013