CASE REPORT

Revascularization of a Nonvital, Immature Permanent Tooth Using Amniotic Membrane: A Novel Approach

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Abstract

Aim: To evaluate the clinical and radiological results of a revascularization treatment done in a nonvital, immature permanent tooth using the amniotic membrane.

Case description: A 12-year-old boy reported with a complaint of pain in the lower left back tooth since 1 month due to dental caries. On clinical examination, the mandibular left second premolar was tender on percussion and discolored due to dental caries. Radiographic examination of the same tooth revealed open apex, thin root dentinal walls, and periodontal ligament widening. An access opening was prepared, necrotic pulp extirpated completely followed by thorough irrigation. After drying the canal, closed dressing with the 3-mix antibacterial paste was given for 15 days. After 15 days, the paste was removed and the amniotic membrane was placed inside the canal extending 1 mm beyond the apex and mineral tri-oxide aggregate was placed over this followed by sealing of the cavity. The patient was followed up at 1, 3, and 6 months for clinical and radiographic evaluation.

Conclusion: After 6 months, clinically the tooth found asymptomatic. Radiographic examination showed continued root elongation, closure in the periapical opening, thick root dentinal walls with narrowing of the canal space, and normal periradicular architecture.

Clinical significance: Amniotic membrane can be used as a scaffold for revascularization in nonvital immature teeth.

Keywords: Amniotic membrane, Immature teeth, Pulp necrosis, Revascularization.

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Introduction

Dental caries or trauma to the permanent teeth leading to pulp necrosis and cessation in root development is the most commonly seen clinical problem in younger children. Arrest in root growth makes the tooth with open apex, fragile root dentin walls, and poor crown–root ratio which all together pose a great challenge in the management of such teeth.[1] Revascularization is the proven therapy in the treatment of immature, necrotic teeth and which needs three pivotal components like signaling molecules, stem cells, and a physical scaffold for its success.[2] Different authors have used various scaffolds like natural blood clot,[3,4] platelet-rich plasma,[5] or platelet-rich fibrin[6–8] for a revascularization procedure. However, these physical scaffolds are found with various pitfalls and contradictory issues.

Amnion is a membrane surrounding the amniotic sac that has been widely used in the field of tissue engineering as it satisfies the biological properties of scaffold material.[9,10] Literature review showed no data pertaining to the application of amniotic membrane for pulp revascularization therapy. Therefore, the aim of this paper is to present a case of revascularization done in an immature, nonvital premolar tooth using amniotic membrane as a new scaffold material.

Case Description

A 12-year-old boy reported to the Department of Pedodontics and Preventive Dentistry complaining of pain in the lower left back tooth since 1 month. On intraoral examination, the mandibular left second premolar had a deep caries involving pulp with mild discoloration and the tooth was tender on percussion. The medical status was noncontributory. The tooth did not respond to cold and electric pulp tests and also exhibited deep periodontal pocket on the distal side. On intraoral periapical radiographic examination, the same tooth revealed deep intrabony defect on the distal side. The tooth also showed an immature root, thin dentinal walls, and open apex with a widening of the periodontal ligament space. Finally, based on clinical and radiographic findings, the diagnosis of the class IV fracture with apical periodontitis was made and treatment of performing a regenerative endodontic treatment using the amniotic membrane was planned. Parents were informed about the treatment protocol and written informed consent was obtained. The protocol and the consent form were approved by the Institutional Ethics Committee (College of Dental Sciences, Davangere, India) to perform the treatment.

Local anesthesia was given using 2% lidocaine with 1:100,000 epinephrine and a rubber dam was applied. Access opening was prepared using a round endo access bur (Dentsply Maillefer, Switzerland) and complete pulp extirpation was done, finally, the canal was copiously irrigated using 10 mL of 5.25% of sodium hypochlorite solution (Rasayan Laboratory, Mumbai, India). The working length was determined using a size 30 k file (Mani, Japan) and the canal was dried with paper points. An equal proportion

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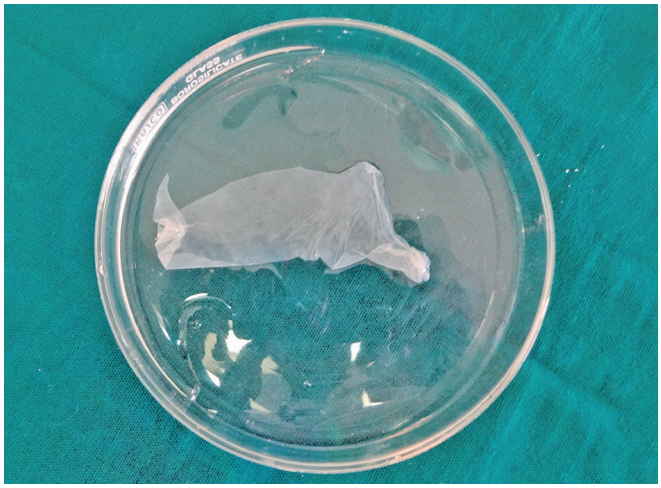
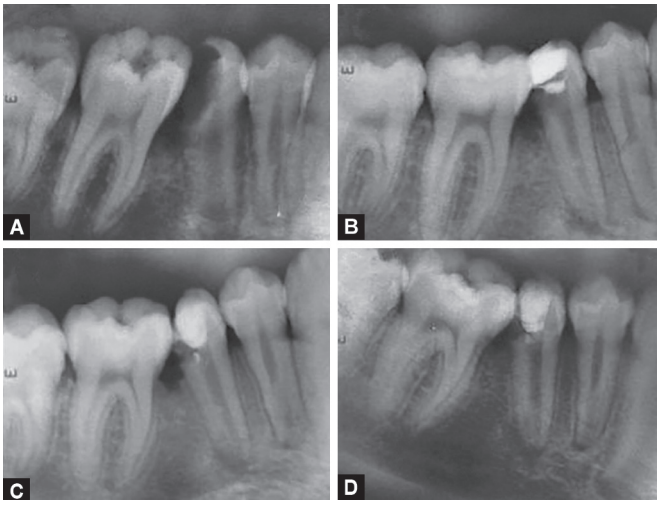


Fig. 1: Picture of amniotic membrane



Figs 2A to D: (A) Preoperative radiograph of 34 showing an open apex, thin root dentine walls, and widening of periodontal ligament space; (B) One-month follow-up showing favorable closure of root apex, regression in the periodontal ligament widening; (C) At the third-month follow up, tooth showing continued root elongation, periapical closure, and thickened dentin walls; (D) At the sixth-month follow-up, tooth showed excellent root elongation, complete closure of the periapical opening, and obliteration of root canal space, thickened root dentin walls and normal periradicular anatomy

of ciprofloxacin (Cifran 500 mg, Ranbaxy Laboratories Ltd., Mumbai, India), metronidazole (Metrogyl 400 mg, J.B. Chemicals, Mumbai, India), and minocycline (Minoz 50 mg, Ranbaxy Ltd., Mumbai, India) was taken, mixed with distilled water, and the paste was placed into the canal to a depth of 1 mm short of the apex using an endodontic plugger (Mani, Japan). The access opening was temporarily restored with Cavit (3M ESPE, Germany) and the patient was recalled after 1 week and the tooth was found asymptomatic. The access cavity was reopened under rubber dam isolation and the canal was thoroughly irrigated using the saline solution to remove the antibiotic paste. Finally, the canal was dried with paper points. The amniotic membrane was obtained (Fig. 1) and placed inside the canal and pushed 1 mm beyond the working length and coronally up to the level of cementoenamel junction (CEJ) using an endodontic hand plugger (Dentsply Maillefer, Switzerland). A thick paste of white mineral trioxide aggregate (MTA) (Pro Root MTA; Dentsply, Switzerland) was placed directly over the amniotic membrane (AM) followed by a wet cotton pellet and the tooth was temporarily restored using the Cavit. The patient was recalled after 1 day to remove the cotton and the access cavity was restored using the glass ionomer cement [GIC (Universal Restorative, Tokyo, Japan)]. The patient was recalled every 1, 3, and 6 months for clinical and radiographic evaluation. After 6 months, clinical examination revealed normal responses to percussion and palpation with decreased pocket depth. The tooth did not respond to cold and electric pulp tests. On radiographic examination, the tooth showed continued root elongation, closure in the periapical opening, thick root dentinal walls with narrowing of the canal space, and normal periradicular architecture (Fig. 2).

Discussion

The traditional method of revascularization as mentioned by Banche and Trope in 2004[2] involves mechanically, irritating the periapical tissues to obtain a fresh blood clot into the canal after complete disinfection of the canal.[1] However, this treatment option is more painful procedure, especially in pediatric patients. In addition, obtaining fresh blood clot is not always possible and there is enough evidence to support this drawback.[11,12] Although various biologically based scaffold materials like platelet-rich plasma or platelet-rich fibrin, and collagen have been used with successful results, each one documented with their own disadvantages. Therefore, there is a quest for the best approach which leads to the development of the amniotic membrane in the arena of tooth tissue engineering.

The amnion is a membrane developing from the fetal tissue consisting of three important layers such as an epithelial layer, a basement membrane, and an avascular mesenchyme.[13] It does not contain nerves, lymphatics, or muscles and could be easily separated from the underlying chorion.[14,15] Human-derived placental tissues are rich in regenerative cytokines and have been studied in randomized clinical trials and showed potential for healing of chronic wounds.[9,15–18] This membrane has been extensively used in the field of tissue engineering because of the favorable biological properties for being a scaffolding material.[19–22]

The first usage of the fetal membrane was done by Davis in 1910 for the transplantation of skin. As the technology in processing and storage methods improved over the time, the use of the amniotic membrane revolutionized in various fields of medicine like for reconstruction of the bladder and the vagina, in the treatment of burns, tympanoplasty, and arthroplasty.[15,23] Pertaining to dentistry, this multipurpose membrane has been used extensively in the field of periodontics,[18] prosthodontics,[24] and oral and maxillofacial surgery.[25–27] However, there is an absence of documentation regarding the use of this membrane as a scaffold matrix for the regenerative endodontic procedures. This tempted us to use the amniotic membrane in revascularization of the necrotic premolar of the patient described here.

In contrast to previous studies,[2–8] we used the amniotic membrane as a scaffold material for revascularization of the pulp tissue in a necrotic, immature tooth as it is a therapeutic potential for soft tissue repair and hard tissue regeneration. It contains many key proteins such as laminin, proteoglycans, fibronectin, collagen types IV, V, and VII, and glycosaminoglycans.[9,16,28] It contains a variety of cytokines like vascular endothelial growth factor, epidermal growth factor, fibroblast growth factor, platelet-derived growth factor, and transforming growth factor beta.[16,21] Another advantageous property of this tissue as an ideal physical scaffold is that it not only provides a matrix for cellular migration and proliferation but also encourages soft tissue healing.[9,26] Moreover, it is a nonimmunogenic,

reduces inflammation, has antibacterial properties, reduces pain at the site of application, and acts as natural biological barrier.[10,21,29] All these properties together make it an interesting biological approach for the application in regenerative endodontic therapy.

On radiographic evaluation, the necrotic premolar treated with the amniotic membrane showed excellent root lengthening, continued thickening of dentinal walls, and regression in the periodontal widening and favorable periapical closure after 6 months. Reasons for this success could be attributed to the presence of angiogenic growth factors retaining biological activity, promotes amplification of angiogenic cues by inducing endothelial cell proliferation and migration and by upregulating the production of endogenous angiogenic growth factors by endothelial cells, and can support the formation of blood vessels.[30] In addition to this, Chen et al.[9] also showed that the amniotic membrane matrix is capable of providing a preferential environment for driving the osteogenic differentiation of human dental apical papilla cells with proven stem cell characteristics. This membrane potentiates the induction effect of osteogenic supplements such as ascorbic acid, β-glycerophosphate, and dexamethasone and enhances the osteogenic differentiation of apical papilla cells. Therefore, as no bleeding was induced in the present case, we can firmly state whatever the further root growth occurred is due the presence of amniotic membrane within the canal.

From this case, it is evident that an amniotic membrane is a promising pulp care therapeutic matrix with the potential to promote revascularization and tissue healing within poorly vascularized immature, nonvital teeth. This case paves a ray for further researches for its application in regenerative endodontic treatment. However, a large number of clinical trials exploring the potential of this scaffold are highly essential in comparison with other scaffold materials before to testify it a boon for regenerative endodontic therapy.

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