Management of immature necrotic permanent teeth with regenerative endodontic procedures — a review of literature

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Abstract

Immature necrotic permanent tooth presents a distinctive challenge for the endodontist. Various treatment modalities have been employed to create hard tissue barrier at the apex, which includes non-vital pulp therapy with calcium hydroxide, apexification with mineral trioxide aggregate, pulp revascularisation and regeneration. Regenerative endodontics is a novel modality which involves physiological replacement of the damaged structures of tooth like dentin, root and cells of the pulp-dentin complex. Numerous published case reports have revealed increased dentinal wall thickness, continued root development and apical closure, but there is still lack of sound scientific evidence regarding histological nature of the type of tissue. The current literature review was planned to summarise the evidence regarding the treatment of immature necrotic permanent teeth by regenerative endodontic procedures.

Keywords: Pluripotent stem cells, Mineral trioxide aggregate, Nonvital tooth, Platelet richplasma, Calcium hydroxide, Apexogenesis.

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Introduction

Permanent immature teeth with necrotic pulp and periapical disease is a constant problem and area of keen interest for endodontists.[1,2] Disinfection of root canal space is difficult to achieve in these teeth with endodontic files using standard protocol.[1,2] Another difficulty arises during root canal filling due to lack of apical barrier in open apex and its impingement on periodontal tissues.[1,2] Even if these challenges are faced and sorted out, the roots of these teeth are very thin that constitute a high risk of fracture.[3-5] Various treatment modalities have been described in literature to create hard tissue barrier at the apex, which includes non-vital pulp therapy with calcium hydroxide, apexification with mineral trioxide aggregate (MTA), pulp revascularisation and regeneration.[1,3,6]

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The traditional non-vital pulp (NVP) therapy with calcium hydroxide (Ca(OH)2) has been studied largely and is reported to have a good outcome.[7] However, there are certain limitations associated with this technique.[5] The principal drawback is the long duration of about 6 and 18 months required for the formation of hard tissue apical barrier and required follow-ups every 3 months to check the progression of barrier formation.[8,9] Patient compliance is extremely important for the completion of the procedure. This technique also tends to decrease the fracture resistance of the root dentine.[4,5] Thus there is always a possibility of root fracture before hard tissue formation.[2,10,11] Kahler et al. recently reported that tendency to root fracture is more related to the stage of root development rather than long-term Ca(OH)2 use.[12] Apexification with MTA has also gained popularity among clinicians which is relatively easy and less time-consuming, but lack regenerative capability and long-term survival is also guarded due to reduced fracture resistance.[13,14] Conventional root canal therapy (RCT) and apexification can only provide apical barrier, but possibility of re-infections and tooth fractures are very undesirable and disappointing for patients and practitioners.[15]

Recently, regenerative endodontics has gained significant interest in the field of endodontics.[16] Considerable research towards successful regeneration of pulp-dentine complex (PDC) is progressing which emphasises on replacement of damaged structures such as dentine, PDC cells and root structures.[17] Tissue regeneration and engineering is the most thought-provoking part of a tissue repair/regeneration programme because of pulp functional importance.[18] The regenerated tissue should be vascularised, innervated and possess the ability to generate new odontoblasts which can produce new dentin matrices that later become mineralised. The concept of regeneration became popular when Banchs and Trope in 2004 treated immature necrotic and permanent teeth with new procedure and termed it 'revascularization'.[19] Pulp revascularisation was an important step by the endodontic community on its path to exploring avenues of pulp and dentin regeneration.[20,21]

Table: Case series/reports published from 2004 till 2017.

Author	Sample (Teeth)	Intracanal Medication	Scaffold	Pulpal Space Barrier/Restoration	Recall	Outcome
Banchs & Trope (2004)[19]	1	Triple- antibiotics	Blood Clot	MTA/Resin	2 years	Hard tissue barrier
Chueh & Huang (2006)[23]	2	Calcium hydroxide	None	Amalgam	7 months to 5 years	Hard tissue barrier
Shah et al.(2008)[24]	14	Formo-cresol	Blood Clot	Glass ionomer	24 months	No hard tissue barrier
Reynolds et al.(2009)[25]	2	Triple-antibiotics	Blood Clot	MTA/composite	18 months	No hard tissue barrier
Thomson & Kahler (2010)[26]	1	Triple-antibiotics	Blood Clot	MTA/glass ionomer/composite	18 months	No hard tissue barrier
Petrino et al.(2010)[3]	1	Triple-antibiotics	Blood Clot	MTA/Resin	08 months	Hard tissue barrier
Torabinejad &Turman, (2011)[20]	1	Triple-antibiotics	PRP**	MTA*/Cavit/amalgam	5½ month	Root Thickening/lengthening
Iwaya et al.(2011)[27]	1	Calcium hydroxide	None	GuttaPercha/ Composite resin	30 months	Hard tissue barrier
Dabbagh et al.(2012)[28]	18	Triple antibiotics	Blood Clot	MTA*/Resin	24 months	Root lengthening and elongation
Martin et al.(2013)[29]	1	Triple-antibiotics	PRP**	MTA/Resin	12 months	Root Thickening/lengthening
Nagata et al.(2014)[30]	23	Triple-Antibiotic/Calcium hydroxide, chlorhexidine	Blood clot	White MTA/ Composite resin	9-19 months	Root Thickening/ lengthening/apical closure
Bezgin et al.(2015)[31]	2	Triple-Antibiotic	PRP**	MTA	12 months	Root Development/apical closure
Nosrat et al.(2015)[32]	2	Triple-antibiotics	Blood clot	MTA*/Resin	4 months	Root Development/apical closure
Bakhtiar et al.(2016)[33]	4	Ciprofloxacin, Metronidazole & cefaclor	PRF***	Bio-dentine	12 months	Root Development/ apical closure
Timmerman (2017)[34]	1	Calcium hydroxide	Blood clot	MTA*/Resin	3 years	Root Development/apical Closure

*MTA- Mineral trioxide aggregate
** PRP- Platelet rich plasma
***PRF- Platelet rich fibrin.

To treat immature necrotic teeth, we have to understand the pathogenesis and various factors that need to be considered before initiating treatment. Immature necrotic permanent tooth results in absence of Hertwig's epithelial root sheath and bacterial colonisation in pulp tissue.[22] Factors that need to be considered include age of the patient, apical diameter, root canal disinfection, and antiseptic irrigants. Young patients have better prognosis due to strong immune defence mechanism and open apex allowing sufficient blood supply.

Multiple case reports /human studies on successful regenerative endodontic procedure have been conducted[19,23,34] (Table), but the current evidence regarding regenerated tissue and regenerative protocol still remains debatable.

The view of many authors regarding pulp regeneration is that it is not possible without revascularisation or angiogenesis and considered incomplete without the formation of an odontoblastic layer that lines the dentin surface. Pulp revascularisation only consists of re-establishment of vascularity in the pulp, but not necessarily the repopulation of odontoblasts that align on the dentin surfaces. Pulp regeneration is not completed without the development of nociceptive, sympathetic, parasympathetic nerve fibres and perhaps most importantly stem cells that aid in replacement of pulp cells in the regenerated pulp.

Thus, it can be concluded that pulp revascularisation is the induction of angiogenesis in an endodontically treated root canal, and pulp regeneration is pulp revascularisation plus the restoration of functional odontoblasts and/or nerve fibres.[6]

Case reports and case series have reported successful outcome of regenerative endodontic treatment in young immature necrotic teeth.[27,35,36] The stage of root development also determines the most suitable treatment option. Teeth at stages 1,2 and 3 (<half, 1/2, 2/3 root formation and open apex respectively) benefit from regenerative endodntic treatment, but teeth at stage 4 (almost complete root formation) can be treated by both regenerative endodontics and MTA apical plug.[37] Teeth with 0.5mm to 1mm apical diameter has the highest success rate.[38] Persistent infection damages the stem cells and halts the process of repair and regeneration, therefore complete disinfection is important for pulp tissue regeneration.[39] Root canal disinfection can be effectively achieved using passive and active ultrasonic irrigation, and negative pressure irrigation.[39]

It has been reported that triple antibiotic paste and concentrated hypocholite result in stem cell damage and ultimately regeneration.[40-43] Besides these, extent of

apical periodontitis, trauma, compliance, patient age and stage of root development also affect the process of regenerative endodontics.[44,45] Regenerative endodontics takes into account the principles of tissue engineering and regenerative medicine which require correct three-dimensional (3D) assembly of dental stem cells, 3D scaffolds and growth factors to form a functional and useful PDC.[18,46]

Dental Stem Cells

The stem cell has the ability of continuous division and production of progeny cells that can differentiate into numerous other cell and tissue types.[17] They are either foetal / embryonic or postnatal/adult.[15] Stem cells are classified into totipotent, pluripotent and multi-potent.[47] Pluripotent stemcells have the ability to become specialiszed cells from all three germ layers, whereas multi-potent cells differentiate only into specialised cells of the tissue of origin.[47] To regenerate a tissue, the best stem cells are embryonic stem cells, but their source is controversial and can raise ethical issues.[47]

Postnatal stem cell sources include variety of human tissues including oro-facial tissues.[18] An important requirement for regeneration of pulp tissues is to obtain stem cells that can differentiate into odontoblasts.[18] All stem cells involved in odontogenesis are ectomesenchymal in origin with the exception of ameloblast progenitor cells.[18] Postnatal mesenchymal stem cells can be classified as stem cells derived from dental tissues (dental pulp, periodontal ligament etc.) and extra-dental sources (bone).[18]

Five types of postnatal mesenchymal stem cells have the reported ability of differentiation into odontoblast-like cells which include stem cells of human exfoliated deciduous teeth (SHED), dental pulp stem cells (DPSC), stem cells of the apical papilla (SCAP), bone marrow-derived mesenchymal stem cells (BMMSC) and dental follicle progenitor cells (DFPC).[48-53]

Scaffold

Scaffold is a 3D structure used in many tissue-engineering applications. When scaffold is seeded with stem cells, they can proliferate and differentiate into new tissues that ultimately replace the scaffold.[54,55] Ideal scaffold must be bio-compatible, sterilisable, non-cytotoxic, should not evoke any inflammatory response, remain stable, and provide cellular support and vascularisation. It should have an inductive ability with added growth factors and morphogens for a more rapid cellular attachment, proliferation, migration and differentiation into a specific tissue.[55]

Currently many regenerative endodontic procedures (REPs) have made use of dentine and the autologous platelet concentrates (blood clot [platelet-rich fibrin-PRF]or platelet-rich plasma [PRP]) to serve as scaffold.[20,56,57] They are believed to induce stem cell proliferation and differentiation from the apical papilla or other pulp cells because of the presence of growth factors.[58] However, many animal studies have highlighted a common feature that the new pulp-like tissue after revascularisation using either PRP alone or in conjunction with pulp stem cells was devoid of odontoblastic cell layer so this tissue could not be considered true pulp.[59] Similar findings emerged from other pre-clinical studies in which odontoblasts were not identified, even if many pulp tissue elements were histologically detected as fibroblasts, blood vessels and collagen.[59]

Massimo et al. in a systematic review concluded that there is no current protocol to achieve a true regeneration of the necrotic pulp tissue using autologous platelet concentrates either in immature or mature teeth and the benefit of the use of PRP to achieve predictable regeneration of the pulp is still unclear.[57]

Growth Factors

Growth factors are the proteins which bind to receptors on the target cell and act as signals that modulate cell behaviour by inducing cellular proliferation and/or differentiation, chemotaxis, angiogenesis and neuronal growth. Growth factors recruit stem/progenitor cells from their perivascular or other niches to replace the damaged cells by differentiating into a specific cell phenotype and proliferation in the area of injury.[17,60] Key growth factors in pulp and dentin formation include transforming growth factor-beta, bone morphogenetic protein and fibroblastic growth factor. Recent REPs focus on utilisation of growth factors derived from dentine and platelets.[20] Studies have reported that dentin can be considered a reservoir of growth factors and other bioactive molecules that, when released, play significant role in repair and regenerative procedures.[61-63]

Smith et al. reported that there are various bioactive molecules, including cytokines, growth factors and matrix molecules, that are present in dentin and pulp which are believed to promote reparative and regenerative events after injury.[62] Bioactive molecules released by the various tissue preparation agents, irrigants like Ethylenediaminetetraacetic acid (EDTA), medicaments and materials commonly used in endodontics influences regenerative events, including chemotaxis, differentiation of odontoblasts, mineralisation, angiogenesis and neurogenesis.[54] Thus, endogenous bioactive molecules

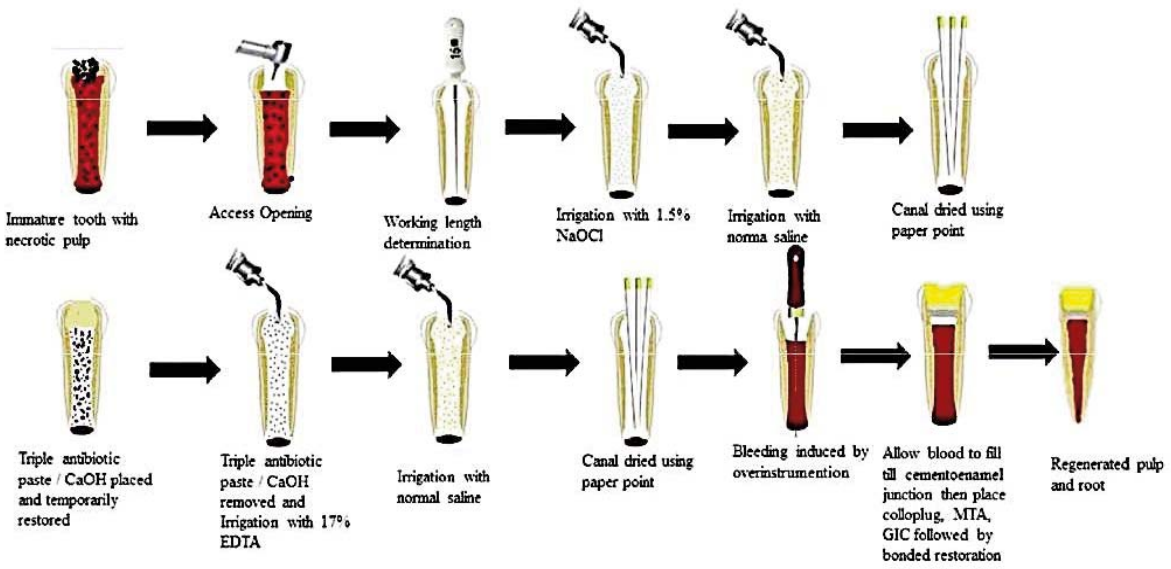

Figure: Protocol of Regenerative Endodontics.

can be targeted to promote regeneration only if the canals are adequately disinfected, which is a pre-requisite for regeneration/repair.

The recommended protocol by the American Association of Endodontics (AAE) (Figure) for regenerative endodontics require initial passive irrigation with 1.5% sodium hypochlorite (NaOCl).[3,19,64-66] Various concentrations of NaOCl has been reported in literature such as 1.25%, 5%, 5.25% and 6% and 2% and 0.12% concentration of chlorhexidine.[1,3,19,65,67-70] At first appointment, slow irrigation with 1.5% NaOCl and then saline, placing the irrigating needle at a distance of 1mm from the apex followed by dressing with triple antibiotic paste (TAP) (ciprofloxacin + metronidazole + minocycline) ensuring that it remains below the cementoenamel junction (CEJ) so as to minimise the risk of staining.[66,71] At second appointment, removal of this paste is done only after resolution of signs and symptoms clinically, bleeding is induced in the canals afterwards using sterile file by over-instrumentation followed by placement of CollaPlug/CollaCote barrier at the orifice. Final step is sealing of canal orifice using MTA followed by permanent restoration with crown.[3,19,71] Follow-up is generally done after 3 months clinically and radiographically to check for resolution of signs and symptoms and increase in length and thickness of the root usually after 12-24 months post treatment.

Limitations of Regenerative Endodontics

Although regenerative endodontics is a great modality to manage immature necrotic teeth, there are limitations to REPs, which include the risk of dental crown-staining induced by materials like TAP/ minocycline and MTA. Clinicians are required to have adequate knowledge to deal with these technical challenges. Use of epinephrine-containing local anaesthetics is a well-known cause of insufficient bleeding; this can be dealt with by using mepivacaine (3%) as an alternative during the second visit before lacerating the apical papilla. However, Hertwig's epithelial root sheath must be protected by avoiding extension of instrument laterally or circumferentially because it directs the shape and continued development of root.[72]

Another common complication that arises is inadvertent staining of the tooth due to exposure to minocycline or TAP used as intra-canal medication. Various strategies have been advised to prevent staining which include sealing of dentinal tubules by bonding agents, use of double antibiotic paste eliminating minocycline or Ca(OH)2 use.[71-73] Use of MTA and other barrier materials are also believed to cause staining, especially when placed above the CEJ.[74] Replacing minocycline by other antibiotic as cefaclor or amoxicillin is another strategy to avoid discolouration. Staining can be prevented by placing the MTA below the CEJ or in the middle third of the canal, use of other biomaterial which has inherent bioactive property like Biodentine, while another approach favours the use of CollaPlug (Zimmer Dental Inc., Warsaw, IN) over the blood clot.[3,33,75]

One major limitation of REP is the lack of the viability of stem cells of apical papilla (SCAP). Pre-existing periapical lesions can damage SCAP, and, similarly, use of NaOClandintra-canal medication at high concentration can also impair SCAP. The in-growth of periodontal tissues (i.e: cementum, periodontal ligament, and bone) into the root canal are believed to be the major reasons for repair instead of regeneration.[76] Unpredictable root maturation is a concern reported in studies.[77,78] but studies have also reported resolution of periapical periodontits in 91% cases, apical closure in 80% and root development in 80% cases.[20,73]

Conclusion

Although the current research in regenerative therapy is very promising, complete biological regeneration of periodontal and endodontic tissues is not yet predictably obtained because of the histological nature of the regenerated tissue which suggests that REPs promote guided-endodontic repair (GER) instead of 'true regeneration' of a PDC. The development of regenerative endodontics may eliminate the need for more complex procedures, like extraction and implant replacement. Further research and randomised clinical trials (RCTs) are needed to develop strong scientific evidence regarding this procedure.

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