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### Case Report

#### Regeneration of Dental Pulp: The Treatment of Immature Necrotic Permanent Teeth

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#### ABSTRACT

Regeneration of the dental pulp like tissue in young permanent teeth with necrosed pulp tissue has provided newer opportunities of treatment. Regeneration of dental tissue is possible due to protheraof prolipotent dental stem cells pre-existing in a properly disinfected root canal system in biological condition having growth factors and scaffold medium. Blood clot formed in the root canal system following intentional over instrumentation acts as an ideal scaffold for the proliferation on stem cells. The present case report presents a case of a 15 year old female patient presenting with a draining sinus in reference to 11 treated with platelet rich fibrin for the regeneration of the dental pulp tissue.

**Key words:** Regenerative Endodontics, calcium hydroxide, tissue engineering, bleeding induction, Biodentin.

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#### INTRODUCTION

Cutting edge dental treatments has moved for simply getting the side effects a greater amount of natural method of consideration, rendering organic substitution of natural tissue. The objective of regenerative dentistry is to prompt organic substitution or production of a natural domain for the recovery and additionally age of dental tissues and their supporting structures.<sup>[1]</sup> Regenerative endodontics is a naturally thought system intended to supplant the lost hard and delicate structures of the tooth i.e dentin, root structure and the structures of the mash dentin complex subsequently permitting the improvement of root length and increment in the thickness of dentinal dividers making the tooth less helpless against fractures.<sup>[2]</sup> Immature necrosed permanent tooth are most suitable tooth for the treatment.<sup>[3]</sup> Mesenchymal stem cells are conveyed through the open apex into the root space of necrosed immature teeth after clinical regenerative method results to revascularization of

the teeth.<sup>[4]</sup> Permitting the host cell homing to shape new tissues in the root space <sup>[5]</sup>.Regenerative endodontics depends on the standards of tissue designing which deals with the right blend of spatial gathering and dynamic cooperation between particular stem cells, development factors and scaffold to shape a pulp dentin complex.<sup>[6]</sup> Clinically regenerative endodontics can be divided in following steps<sup>[7]</sup>:

- Disinfecting the root canal space with minimal or no preparation using a suitable intracanal medicament.
- Treatment with blood clot or autologous fibrin matrix implantation i.e platelet rich plasma or platelet rich fibrin.

#### CASE REPORT

A fifteen year old female patient reported with the chief complain of pus discharge from gums in upper front tooth region for last 20 days.On further scrutinizing a

background marked by injury was given by the parents 6-7 years ago. For which they had given some analgesics to treat the pain. Dental, restorative and family history was non-contributing. Intra oral examination sinus was present and with no related tenderness on percussion. Intra oral periapical radiograph showed radiolucency of around 2mm in measurement in connection to 11. Apical closure was as yet occur in 11. Vitality test by hot test, cold test and electronic pulp tester was performed and all were non responsive. Taking into consideration the age of the patient, pulp vitality status and non closure of apical foramen pulp Regenerative Endodontic Procedure was considered over apexification as regenerative treatment will enhance the strength of root by formation of dentin.. The patients parents were explained about the treatment (along with its limitations) to which they agreed. A written consent form was signed by the parent of the patient.

On the 1<sup>st</sup> appointment access cavity was prepared in 11 under rubber dam isolation the working length was determined utilizing electronic apex locator ( Root ZX II, J Morita, Japan) and checked on an intra oral periapical radiograph. Circumferential filling was finished with an ISO 20 no. K-file upto the decided working length to expel all the necrotic tissue alongside copious irrigation with 1.5% Sodium Hypochlorite for 5 minutes with a side vented needle followed by 20ml of distilled water for 5 minutes. This was in agreement to clinical convention by AAE for regenerative procedure.<sup>[3]</sup> The needle tip was put 2mm short of the working length for achieving proper disinfection of the canal avoiding peri apical extrusion of the irrigants. Root canal space was dried with sterile paper point and Calcium hydroxide and iodoform was placed as an intracanal medicament. A pellet of sterile Teflon tape was placed and access opening was restored with restorative Glass Ionomer Cement (Fuji II).

The sinus opening had reduced to approximately half of its original size on the second appointment after a week . Re-entry into the tooth was made and intra canal medicament was removed using normal saline and a ISO 15 no. K-file making minimal contact with the canal walls. Followed with through irrigation with 1.5%, 20ml Sodium Hypochlorite for 5 minutes with a side vented irrigation needle followed by 20ml of normal saline for 5 minutes. Canals were dried with sterile paper points and intra canal medicament was repeated .A pellet of sterile Teflon tape was placed on the orifice and access cavity was restored with restorative Glass Ionomer Cement (Fuji II).

Sinus had completely resolved on the third appointment 7days after the second appointment. Re-entry into the tooth was made and intra canal medicament was removed same as on second appointment. Followed with through irrigation following same protocols followed on 2<sup>nd</sup> appointment. Canals were dried with sterile paper points. 10 ml venous blood was withdrawn from the patient and centrifuged in a centrifugal machine(Remi Model R8C) at 2500 rpm for 12 minutes to obtain autogenous platelet rich fibrin (PRF). A

sterile ISO 15 No. file was inserted 2mm beyond the working length to induce fresh bleeding the opening was covered with cotton pellet and waited till clot formation started . PRF was cut in small pieces and introduced into the canal with a help of a hand plugger with minimal apical pressure avoiding any rupture of the PRF simultaneously placing the PRF in the apical region. A bi dentine bridge was placed on the coronal side of the access cavity and the cavity was restored using restorative GlassIonomer Cement (Fuji II).

The patient was advised follow up visits 1 week, 1 month and thereafter every 3 months . The patient did not show any symptoms on percussion and palpation tests , the teeth showed negative response to heat and electronic pulp tester (EPT) on the 1 week and 1 month follow up visits. On the 3<sup>rd</sup> month follow up did not show any symptoms on percussion and palpation tests , the teeth showed negative response to heat and electronic pulp tester (EPT). Intraoral periapical radiograph revealed thickening of the dentinal walls. On the 9<sup>th</sup> month follow up visit the patient did not show any symptoms on percussion and palpation tests , the teeth showed negative response to heat and electronic pulp tester (EPT). Intraoral periapical radiograph revealed continued thickening of the dentinal walls, reduction in periapical radiolucency and apical closure.

On the 1yr follow up of the did not show any symptoms on percussion and palpation tests , the teeth showed negative response to heat testing , but a positive response was present on cold testing and electronic pulp tester (EPT) at reading 8 suggestive of the pulp tissue regeneration and regain of sensibility. Intraoral periapical radiograph revealed continued thickening of the dentinal walls, reduction in periapical radiolucency and apical closure. Patient was advised follow up recalls every 6 months.



9<sup>th</sup> month follow up



6<sup>th</sup> month follow up



3 month follow up



Immediate placement of PRF



Reconfirmation of working length after removal of intracanal medicament



1<sup>ST</sup> Month follow up

### DISCUSSION

Regeneration of the pulp tissues in an immature permanent teeth of an young adult has opened new horizon on the longevity of the tooth against fractures by substituting the hard tissues such as dentin , root structure and cells of the pulp dentin complex leading to increase in root length and apical closure. The present case report demonstrates the potential for revascularization of the tooth with infected root spaces with some pulpal tissues. Widening of the root canal walls and closure of the apical region was seen. The most critical criteria for choice for revascularization are teeths with necrotic pulp with wide opening of the root summit. Kling M et al. demonstrated that if the zenith demonstrated a radiographic opening of 1.1mm the odds of revascularization expanded by 18%.<sup>[9]</sup>

The irrigation regime pursued and use of calcium hydroxide iodoform paste was done as per clinical conventions of American Association of Endodontics for regenerative methodology. The earliest case reports depicting the cleansing convention in the treatment with REPs included disinfecting the root canals with a blend of antibiotic agents.<sup>[10,11]</sup> Notwithstanding, others upheld the utilization of calcium hydroxide.<sup>[12,13]</sup> In a larger part of published cases appear to have utilized a mix of antibiotics as the favored intracanal medicament.<sup>[14]</sup> Nonetheless, these antibiotic pastes have announced cytotoxicity to the survival of SCAP in a fixations equivalent to or higher than 1 mg/mL in vitro examinations. Then again, the disinfection of root channel space with calcium hydroxide advanced the expansion of SCAP.<sup>[15,16]</sup> Calcium hydroxide has additionally been accounted for to build the arrival of development factors from dentine, though antibiotic pastes adversely affected development factor discharge after the utilization of EDTA.<sup>[17,18]</sup>

Plasma rich fibrin (PRF) a second generation blood derivative was used as a scaffold in this case. It contains a lot of growth factors such as PDGF, TGF  $\beta$ 1, and IGF PRF consists of an intimate assembly of cytokines, glycan chains, structural glycoproteins enmeshed within a slowly polymerized fibrin network. These biochemical components have well known synergistic effects on healing processes. Fibrin is the natural guide of angiogenesis. Fibrin constitutes a natural support to immunity.<sup>[19]</sup> Bose R and others, in a review radiographic investigation utilizing a quantitative examination of teeth treated with REPs went on the defensive cured with calcium hydroxide had an altogether more prominent increment in root length than the teeth cured with the mix anti-biotic paste. Anyway teeth treated with the mix antibiotic paste had essentially more noteworthy increments in root canal divider thickness. The arrangement of calcium hydroxide within the root canals appeared to positively influence the result of the treatment.<sup>[20]</sup>

The regeneration of pulp/pulp like tissue relies upon the idea of Tissue Engineering using the dynamic connection between the development factors, foundational microorganisms and platform medium. The presence of different undifferentiated cells having one of a kind separation and duplication potential into more current cells have been recognized in immature teeth. Development factors influence a wide scope of cell exercises including movement, multiplication, separation, and apoptosis of the dental pulp cells, including immature microorganisms. For coronal sealing BIODENTIN is regarded as the material of choice as it gives excellent coronal seal and good results as tissue growth and aesthetic consideration (in accordance AAE protocol for regenerative endodontics dated 04/01/2018) and access cavity was restored glass ionomer cement with beveled margin.

Biodentine is a bioceramic cement in the same family of compounds as MTA. The material is composed of

tricalcium silicate, dicalcium silicate, zirconium oxide, calcium carbonate, calcium oxide and iron oxide. It is mixed with a hydrosoluble polymer and calcium chloride to decrease the setting time.<sup>[23]</sup> Biodentine is an alkaline cement with a pH range of 11.7–12.3.<sup>[24]</sup> This material has also been shown not to be cytotoxic to pulp fibroblasts.<sup>[25]</sup> The dentine bridge formation formed by MTA and Biodentine was found to be comparable<sup>[26]</sup>. Biodentine can also be used as an intracanal barrier over the blood clot.<sup>[27,28]</sup> and has been shown to stain teeth less than MTA.<sup>[29-31]</sup>

In the present accessible literature regenerative endodontic strategies in immature non-vital teeth with acceptance of blood clot and PRF. The blood clot was not used alone as a scaffold as it might cause discomfort to the patient and most importantly amount and extent of blood clot formed is not in control of the operator. So better methodology in autologous fibrin network as Platelet Rich Plasma or Platelet Rich Fibrin in revascularization strategy.

## CONCLUSION

The benefit of root lengthening and widening by formation of cementum and dentin in regenerative endodontic procedure makes it an attractive alternative against traditional apexification procedure for non-vital immature teeth. However, the predictability of such procedure is limited and the type of tissue regenerated has to be studied in more precise details before a conclusion is drawn.

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