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This article describes about a biological approach in the repair of dental pulp and periapical tissues using biomaterials (platelet rich fibrin [PRF] and mineral trioxide aggregate [MTA]) and laser biostimulation. Case 1: Pulpotomy with PRF, MTA, and laser biostimulation. Patient follow-up was done at 1, 3, and 6 months. On 6 months follow-up, the patient was asymptomatic and tooth responded positively to pulp tests. Radiograph revealed normal periodontal ligament space and trabecular bone pattern. Case 2: Apexification procedure using PRF, MTA, and laser biostimulation in 31 and 41. Patient follow-up was done at 1, 3, and 6 months. On 6 months follow-up, radiograph revealed relative root lengthening, thickening of the canal walls in the apical one-third of root, and a healing lesion. Repair and regenerative potential of PRF, an enhanced cellular metabolism with laser biostimulation in combination with the sealing ability of MTA enhances the clinical success outcomes in pulpotomy and apexification procedures.

Keywords: Apexification, Biocompatible materials, Low-level laser biostimulation, Pulpotomy

INTRODUCTION

Biomaterials are native or synthetic polymers that perform as scaffolds for tissue regeneration and hold wide importance in the field of dentistry, drug delivery, cancer treatment, thrombotic diseases, and cosmetic surgery.¹ Based on the reaction of the tissue to the biomaterial, these are classified into three distinct categories: Biotolerant materials, bioactive materials, and bioinert materials.² Various bioactive materials are available in today's time like mineral trioxide aggregate (MTA), bioactive glass, and bioaggregate materials. In regenerative endodontic therapy, an ideal autologous biomaterial for pulp-dentin complex regeneration is platelet-rich fibrin (PRF).³

PRF was first developed by Choukroun et al., in 2001 at France. Studies have demonstrated that the PRF has a very significant slow sustained release of many key growth factors like platelet-derived growth factor and transforming growth factor-β for at least 1 week and up to 28 days, which means that PRF could release growth factors with its own biological scaffold for wound healing process.⁴ Various clinical applications of PRF include, root coverage, bone regeneration, treatment of endo-perio lesions, sinus floor elevation, stabilize graft material in ridge augmentation, socket preservation, stabilize graft material in ridge augmentation, socket preservation, filling cystic cavities, and in various medical fields.⁵

MTA was introduced by Dr. Torabinajed in 1993. It is established as osseo conductive, inductive, and biocompatible material. This material was developed and recommended initially as a root-end filling material and subsequently has been used for pulp capping, pulpotomy, apexogenesis, apical barrier formation in teeth with open apexes, repair of root perforations, and as a root canal filling material.⁵

Low level laser therapy as a clinically well accepted tool in regenerative medicine and dentistry improves healing processes and management of function disorders. Providing direct biostimulative light energy to cells is the...
main target of low level laser therapy. Low intensity laser therapy has favorable results on a variety of pathologic conditions such as pain and inflammation, chondral and fibroblast proliferation, collagen synthesis, and nerve regeneration.\(^6\) Biostimulation laser therapy is one which plays an important role in accelerating the repair process of injured tissues.\(^7\)

It is important to develop biocompatible treatment modalities directed at maintaining pulp vitality and increasing tooth longevity. To increase the success rate, a critical need exists to develop new biologically based therapeutics that reduce pulp inflammation and promote the formation of dental pulp and periapical tissues. This article describes a biological approach in the repair of dental pulp and periapical tissues using PRF, MTA, and laser biostimulation.

**CASE REPORTS**

**Case 1: Pulpotomy with PRF, MTA and Laser Biostimulation**

A 21-year-old female patient reported to the Department of Conservative Dentistry and Endodontics with pain in the lower left posterior region. Patient had a history of sharp pain, lingering response to a cold stimulus. On clinical examination, occlusal deep dental caries was seen on the left mandibular molar (36). On vitality tests, tooth showed responses related to irreversible pulpitis. An intraoral periapical radiograph revealed deep occlusal caries invading the pulp with slight periapical rarefaction and mild condensing osteitis in relation to the distal root (Figure 1). The diagnosis of chronic irreversible pulpitis was determined on the basis of clinical and radiographic assessment. The patient was informed about the treatment modality - coronal pulpotomy followed by PRF and MTA as an alternative treatment to root canal treatment.

After explaining treatment protocol, a written consent was obtained from the patient. In the first visit, tooth 36 was first anesthetized with Lidocaine 2% and adrenaline 1/80000 (Lignox, Indoco Remedies Ltd, Mumbai, India) and isolated with a rubber dam. Meanwhile, PRF was prepared by drawing the required amount of blood into a 10-mL test tube without an anticoagulant and centrifuged immediately using a table top centrifuge (REMI R-8C, Laboratory Centrifuges Mumbai, Maharashtra, India) at 2400 rpm for 12 min. The resultant product consisted of the three layers: Acellular platelet poor plasma at the top of the tube, fibrin clot PRF in the middle of the tube and red blood corpuscles at the bottom of the tube (Figure 2).

After confirming anesthesia using subjective and objective findings, complete caries excavation, and access opening was done. Pulpotomy was performed with a large round bur (Mani, Tochigi, Japan) in a high-speed handpiece with copious saline irrigation; coronal pulp tissue was removed to the level of root canal orifice. Hemostasis was achieved by irrigating the cavity with sterile saline and cotton pellets (Figure 3). The blood clot-free pulpal wound was irradiated with diode laser 810 nm diode laser (DenLase - 810/7, China Daheng Group, Inc., China) at biostimulation mode, 0.3 W for 20 s (Figure 4) and was covered with a small piece of PRF membrane (Figure 5). An approximately 2 mm thick...
layer of MTA (Angelus, Londrina, PR, Brazil) was placed over the PRF (Figure 6). Coronal sealing was done with glass–ionomer cement (Fuji IX; GC Corp, Tokyo, Japan) (Figure 7) and immediate post-operative radiograph was made (Figure 8). Patient recalled after 1 week.

In the second visit (after 1 week), the patient was asymptomatic and the coronal restoration was intact. Radiograph revealed no periapical changes. Patient follow-up was done at 1 (Figure 9), 3 (Figure 10), and 6 (Figure 11) months. On 6 months follow-up, the patient
was asymptomatic and tooth responded positively to pulp tests. Radiograph revealed normal periodontal ligament space and trabecular bone pattern.

**Case 2: Apexification Procedure Using PRF, MTA, and Laser Biostimulation**

A 18-year-old male patient reported to the Department of Conservative Dentistry and Endodontics with pain in the lower front teeth region. Patient had undergone trauma during his childhood, and complains of pain which was mild, continuous, and throbbing type. On extra oral examination, mild swelling was noticed over the chin region. Intraoral examination revealed fracture involving pulp and tooth discoloration in 31 and 41 (Figure 12). There was a pus discharge from the sinus tract opening in relation to 41 (Figure 13). Vitality tests showed non-vital responses in 31 and 41. Periapical radiograph revealed relatively wide canals with open apices in 31 and 41 (Figure 14). A diffuse radiolucency with ill-defined borders of about 10 mm × 7 mm in relation to 31 and 41 was observed periapically. A diagnosis of non-vital tooth with periapical abscess i.r.t 31 and 41 was done. A regenerative endodontic procedure using PRF and laser bio-stimulation was planned for 31 and 41.
In the first visit, access cavity preparation, working length determined, minimal biomechanical preparation, and copious amount of 3% NaOCl and saline was done in 31 and 41. In the second visit, canals irrigated, laser disinfection done with 810 nm diode laser (DenLase - 810/7, China Daheng Group, Inc., China) at 1.5 W, continuous wave and 1mm short of apex in helicoidal motion from apex to the coronal aspect of tooth. This procedure repeated 4 times for 5 s. In the third visit, asymptomatic teeth 31 and 41 were irrigated and dried. Bio-stimulation of the periapical tissue was done using 810 nm diode laser (0.3 W for 20 s, glass fiber tip was adjusted 1 mm short of apex using silicon stopper) followed by placement of the PRF into the periapical tissue and the root canal space with the help of an endodontic plugger. Over PRF, approximately 2 mm thick layer of MTA (Angelus, Londrina, PR, Brazil) was placed and coronal sealing done with Glass–ionomer cement (Fuji IX; GC Corp, Tokyo, Japan) in 31 and 41 (Figure 15). The patient was recalled after 1 week for clinical and radiographic examination.

In the third visit (after 1 week), the patient was asymptomatic, coronal restoration was intact, and radiograph revealed no periapical changes.

Patient follow-up was done at 1 (Figure 16), 3 (Figure 17), and 6 (Figure 18) months. On 1 month follow-up, the patient was asymptomatic and sinus tract opening was completely closed. On 3rd month follow-up, there was a reduction in size of the lesion periapically. On 6 months follow-up, radiograph revealed relative root lengthening, thickening of the canal walls in the apical one-third of root and a healing lesion in relation to 31 and 41. Biomechanical preparation and complete obturation were done in 31 and 41. Post endodontic restoration was done with the light cure composite restorative material.

**DISCUSSION**

In regenerative endodontics, a successful vital pulp treatment requires a good sealant against bacteria, no severe inflammatory reactions, and stable hemodynamic within the pulp. Pulpotomy is a universally accepted treatment for teeth with incompletely formed roots involving pulpal exposure. In permanent teeth, it has been postulated...
that extirpating pulpal tissue and undertaking root canal treatment in many cases is not cost-effective as it is time-consuming and difficult for both patient and clinician. In addition, failure of a vital pulp therapy would not reduce the outcome of future root canal treatment for the tooth.9

PRF organizes as a dense fibrin scaffold with a large number of leukocytes concentrated in one part of the clot, with a specific slow release of growth factors (e.g., transforming growth factor-1β, platelet derived growth factor-α β, and vascular endothelial growth factor) and glycoproteins (e.g., thrombospondin-1) over ≥7 days.3 PRF slows down the blood activation process, which could induce an increased leukocyte degranulation and cytokine release from proinflammatory mediators, such as interleukin (IL)-1β, IL-6, and tumor necrosis factor-α.10 PRF stimulates the proliferation and differentiation of dental pulp cells by up-regulating osteoprotegerin and alkaline phosphatase expression.4 In case of pulpotomy with PRF, the role of PRF in the process of wound healing occurs in 3 stages - biochemical activation, cellular activation, and cellular response.11 The vascular endothelial growth factor released from PRF matrix was primarily responsible for endothelial mitogenic response via extracellular signal-regulated protein kinase activation pathway.12 This could be the reason for the biological repair of irreversible pulpitis with PRF as matrix over the inflamed pulpal tissue.

Root dentine is formed by root odontoblasts differentiated from ectomesenchymal cells in the apical papilla upon receiving inductive signal from the Hertwig’s epithelial root sheath (HERS). Cementum is formed by cementoblasts differentiated from ectomesenchymal cells in the dental follicle after receiving inductive signal from the HERS. In addition, stem cells in the periodontal ligament are also capable of differentiation into cementoblasts. PRF stimulates osteoblasts, gingival fibroblasts, and periodontal ligament cells proliferation as a mitogen. Many growth factors such as platelet derived growth factors and transforming growth factors are released from PRF.13 The properties of this natural fibrin biomaterial thus offer great potential during wound healing, apex development, and bone regeneration. Outcome of apexification procedure in the present report showed Type-1 (increased thickening of the canal walls and continued root maturation) pattern of root repair according to Chen et al.13

Diode lasers can be used for a multitude of dental procedures including management of soft and hard tissues. Near infrared laser light around 810 nm to be one of the most versatile wavelength ranges in diode lasers available to the dentist. Low-intensity laser biostimulation shortens the inflammatory phase of the healing process, speeds up cell proliferation, and enhances neovascularization.14 In endodontics, for an effective disinfection of the root canal, the power should be set in the range of 1-1.5 W.15

A layer of MTA was placed over PRF and the final restoration of glass–ionomer cement was placed immediately. MTA has been introduced for pulpotomy in primary molars and has demonstrated good biocompatibility, excellent sealing ability, and stimulation of healing in the pulpal tissue. MTA was chosen as it is hydrophilic and requires moisture to set, which is a favorable property when there is potential for moisture contamination in the clinical setting. Among the conventional restorative materials, MTA with a combination of GIC is recommended as good coronal sealing materials to prevent microleakage.16 This combination provides a double coronal seal (MTA+ glass ionomer cement) eliminate ingress of microorganisms, creating a stable environment. MTA in its freshly mixed state shows a higher cytotoxicity, which could be due to its high pH.11 Therefore, PRF acts as a protective barrier on the pulp to prevent the direct contact of freshly mixed MTA.

**CONCLUSION**

Repair and regenerative potential of PRF, an enhanced cellular metabolism with laser biostimulation in combination with the sealing ability of MTA enhances the clinical success outcomes in pulpotomy and apexification procedures.

**REFERENCES**


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