CASE REPORT

Equine-derived xenograft and PRF for intrabony defect of localized aggressive periodontitis – A case report

Nirupa Elisetti, Pallavi Samatha Yalamanchili, M. V. Ramoji Rao

Department of Periodontics and Implantology, Drs. Sudha and Nageswara Rao Siddhartha Institute of Dental sciences, Chinnoutpally, Andhra Pradesh, India

Abstract

The 21st century is revolving around new generations of grafts and their regenerative potential in treatment of intra bony defects. Growth prospect of any material potentiates when additive agents of similar function are coordinated together yielding satisfactory clinical outcomes. An equine-derived cancellous bone graft which is thought to have similar efficacy as proven hydroxyapatite crystals for the regeneration of intrabony defects was used for comparison in a localized aggressive periodontitis patient. Platelet-rich fibrin is also used as an adjunct along with the graft to synergize its effect. Clinical outcome was positive with a reduction in probing pocket depth and radiographic analysis also showed significant bone fill in the defect.

Keywords: Aggressive periodontitis, bone graft, platelet-rich fibrin

Introduction

Regeneration is defined as a reproduction or reconstruction of a lost or injured part in such a way that the architecture and function of the lost or injured tissues are completely restored (Glossary of Periodontal Terms, 1992). The dynamic and biological process of wound healing aims at reproduction of new cementum formation with insertion of new periodontal ligament fibers, along with formation of alveolar bone. Melcher’s Concept (1976) of compartmentalization relies on the type of cell which populates the surgical site wins the race and occupies the area. The cells could be epithelial cells, gingival connective tissue cells, periodontal ligament cells, and cells from alveolar bone. The spatial orientation of the cells and their sequence of entry to the disturbed part and their temporal relationship remain rather challenging for regeneration. Over the years, many regenerative techniques have evolved toward achievement of success with relation to development of periodontal tissues flawlessly function able. Grafting still remains gold standard for regeneration; however, advancements in graft material make head way toward perfection. Many challenges have been identified in regeneration of new bone even after focusing on discovery of befitting agents. Incorporation of root bio-modification, principles of guided tissue regeneration (GTR), and application of growth factors has been followed as additive procedures to attain perfection. Nonetheless, understanding the basic cell kinetics and physiology of wound healing drives us toward excellence aiming at meticulous technique sensitive procedures followed. This case report describes comparison of two different bone graft materials used, along with adjunctive use of platelet-rich fibrin (PRF) placed in a three walled bony defect in a localized aggressive periodontitis (LAP) patient.

Clinical Presentation

A 20-year-old systemically healthy male patient reported to the Department of Periodontics and Implantology in 2017 for a routine oral check-up. On clinical examination, a seven millimeter deep periodontal pocket was observed in relation to first molar on both right and left side as measured with William’s periodontal probe [Figure 1c]. Similarly, deep pockets were observed in lower molars and central incisors with pathological migration in relation to upper incisors [Figure 1b]. Very minimal deposits were seen, with no inflammatory changes and recession in gingiva. Anterior deep bite with mild attrition in lower incisors was observed. The teeth were firm in their sockets. Frenal attachment in relation to upper arch was papillary penetrating. Laboratory investigations of complete blood picture, bleeding time, clotting time, HbA1c, and HbsAg

Correspondence:
Dr. Nirupa Elisetti, Department of Periodontics and Implantology, Nirupa’s Dental Zone, Nallagandla, Hyderabad - 500019, Telangana, India. Phone: +91-6309072369. E-mail: nirupasadentalzone@gmail.com

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revealed no abnormalities. Radiological investigations of osteoprotegrin (OPG) [Figure 1a] and a full mouth intra oral periapical radiographs revealed arc shaped radiolucencies in relation to upper molars on both sides and also left lower molar. Bone loss was also observed in relation to both upper and lower anteriors. A diagnosis of LAP was made pertaining to arc shaped radioluencies confined to all molars and incisors with barely noticeable deposits present, along with deep pockets. The treatment with localized open flap debridement, along with placement of graft and PRF membrane and frenectomy, was planned. The patient was informed about the diagnosis and the procedures were explained. He was compliant enough and gave his consent. As a general rule, oral prophylaxis was performed before surgical appointment. Two different bone graft materials were planned on either side (right and left) to observe their regenerative capacity, along with PRF.

**Case Management**

On the day of surgery planned, the patient was asked to have adequate breakfast before coming for the appointment. A solution was made with dispersible tablet of Ketorolac Tromethamine 10 mg and given to the patient as a premedication for pain during the procedure. As planned for the regeneration of defects with bone grafts, along with PRF, 10 mL of blood were drawn from the ante-cubital fossa with a disposable sterile syringe under aseptic conditions. The blood was divided into two equal halves of 5 mL each in two different test tubes without anticoagulant as a requirement for extraction of PRF. They were divided so for obtainment of two PRF membranes to be used on contra-lateral sides [Figure 2a]. Now, both the test tubes were centrifuged at 2500 rpm for 10 min. The acellular plasma was drained out which is the top layer, and then, the fibrin layer in the middle was placed on a sterile piece of guaze to suck the remaining unwanted plasmatic fluids and then squeezed to obtain a PRF membrane [Figure 2b].

The patient rinsed his mouth with 5% povidine iodine solution before the surgery. Local anesthesia of 2% lignocaine with 1 in 100,000 adrenaline was given to the area to be operated. Modified Kirkland flap surgery with sulcular incision was placed and a full thickness mucoperiosteal flap was elevated [Figure 3a] and debridement of granulation tissue was done to render the area infection free. A three wall intraosseous bony defect was perceived on either side. Bone graft used in the upper left quadrant is an alloplastic graft, G-bone graft block (synthetic hydroxyapatite granules) manufactured by Surgiwear, India [Figure 3d and Figure 4a]. Whilst graft used on the contra lateral side was a xenograft, Biogen (bony substitute of equine origin) manufactured by BioTeck, India [Figure 3b and Figure 4b]. Bone grafts were tucked into the defects and shielded with PRF membrane [Figure 3c]. Flap was reassembled and fastened with 3-0 black braided silk.

**Post-operative care**

Antibiotic medication of amoxcillin-500 mg thrice daily for 5 days and an analgesic of paracetamol-650 mg every 8th hourly for 5 days was prescribed. The patient was advised to be on cold and soft diet for 2 weeks and was recalled after 10 days for suture removal. He was instructed not to brush his teeth for 1 week and was asked to rinse his mouth with 0.2% chlorhexidine gluconate solution beginning from 2nd day after surgery. The patient was followed on the same day by phone conversion to verify his comfort level. He responded well with no post-operative swelling, discomfort or fever.

**Clinical outcomes**

A recall visit after 1, 3, 6 and 8 months was followed reinforcing the oral hygiene practices. Reduction in probing pocket depth was observed in both the first and second quadrants at the 8th month follow-up appointment. Similarly, radiographs at 8th month follow-
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Figure 3: (a) Full thickness mucoperiosteal flap reflection done and three wall intraosseous bony defect seen in relation to first quadrant molar. (b) Biogen bone graft material placed in the intrabony defect of first quadrant. (c) PRF membrane placement over the graft material in the first molar region of first quadrant. (d) G-bone graft material placed, along with PRF membrane in the first molar region of second quadrant.

Figure 4: (a) G-Bone graft. (b) Biogen equine-derived bone graft

Figure 5: (a) Eight months post-operative radiograph in relation to first quadrant. (b) Eight months post-operative radiograph in relation to second quadrant.

Figure 6: (a) Frenectomy done in relation to upper teeth. (b) Immediate post-operative. (c) Immediate post-operative on the palatal aspect. (d) Post-operative on 10th day follow-up.

Discussion

Aggressive periodontitis is rapid destruction of periodontal apparatus in an otherwise healthy individual with familial aggregation of cases as defined by Lang et al, 1999. The present case deals with a systemically fit non-smoker male patient with severe bone loss and no family history. Localized aggressive categorization was confinement of disease to specific teeth such as incisors and molars and age of the patient was 20 years (circum pubertal age group). However, serum antibody response to specific infectious agent was not done, which remains a drawback for the case. The presence of local factor (plaque) was almost negligible as proportionate with the amount of destruction present. Pathological migration in the upper anterior region can also be attributed to the deep bite and abnormal frenal attachment. Hence, diagnosis of LAP was made correlating all the positive findings. Open flap debridement with regeneration of the local areas, along with correction of frenum and little grinding of anterior teeth for occlusal correction, was planned.

Thorat et al. investigated the potency of autologous PRF alone in treatment of LAP intrabony defects in 15 patients and concluded with a satisfactory improvement in both clinical (decreased probing pocket depth and gain in clinical attachment) and radiographic bone fill in accordance to the 8 months follow-up of this case. Bajaj et al. also studied utilization of PRF in fifty four intrabony defects of seventeen aggressive periodontitis patients with positive clinical outcome.

PRF membrane, a second generation platelet concentrate discovered by Choukroun et al., has revolutionized regeneration...
concept with platelet concentrates, along with leukocytes in a fibrin mesh work. In a healthy healing wound, clot mechanism stimulates fibrin web with embedded platelets and leukocytes to release cytokines and growth factors stimulating the natural process. The same scenario has been established in PRF obtained without anticoagulant, resulting in retention of cellular integrity, devoid of any cross reacting agents. Leukocyte rich platelet-rich fibrin (L-PRF) concentrates transformed into a membrane also acts as GTR as with its abundance of platelets acting as a scaffold for the defect and, in turn, aids in release of numerous growth factors like platelet-derived growth factor, fibroblast growth factor, and transforming growth factor-β. PRF is a very potential treatment option as an adjunct to bone grafts for regeneration.\(^5\)

Thorat et al. stated that PRF-treated intrabony defects showed radiological improvement in bone fill as compared to controls.\(^6\) Pradeep et al. conducted a study to compare the efficacy of platelet-rich plasma and PRF in 90 three-wall intrabony defects and concluded that both were equally successful pertaining to bone fill of defects.\(^7\) The present case, wherein PRF was used as an adjunct to bone grafts, improved bony trabecular pattern and decreased clinical probing depth with attachment gain at 8 months follow-up on either sides. Pradeep et al. investigated the efficacy of PRF, along with porous hydroxyapatite graft in three walled bony defects and observed improvement in clinical parameters in test group as compared to group with PRF alone without bone graft. Hydroxyapatite graft when placed along with PRF added to capacity of regeneration.\(^8\) This is in agreement to the present case where bone grafts (equine derived and hydroxyapatite) were placed along with PRF membrane which yielded satisfactory clinical results.

Grafts can be autogenous, xenograft (from different species) or alloplast (hydroxyapatite). Autogenous derived from one’s own body is the gold standard. However, alloplastic grafts with their osteoconduction effect of bone apposition also give promising results of bone regeneration. Xenografts such as bovine derived have shown good results in regeneration. Equine-derived grafts were rarely used and efficacy of it needs to be still established. Nevins et al. examined the histological bone regenerative potential of equine-derived bone mineral matrix for maxillary sinus floor augmentation.\(^9\) Histological analysis revealed good interplay of newly formed bone with equine bone graft with no inflammatory changes observed with good clinical bone formation for implant placement. Equine-derived bone graft (BIOGEN) was placed in upper right molar defect and synthetically derived hydroxyapatite (G-Bone) in the left molar defect. Regeneration of bone and bone fill was equally satisfactory on both sides. Regeneration of periodontal tissue depends on the first cell migrating into the defect area and clinically effective results could be achieved only when periodontal ligament fiber cells repopulate the area prior to any other occupation. Chang et al. determined the effect of PRF on periodontal ligament fibroblasts by measuring the expression of osteoprotegrin, alkaline phosphatase, and phosphorylated extracellular signal regulated protein kinase and showed their increased expression signifying the positive effect of PRF on bone regeneration.\(^{10}\) This case report has a different presentation of localized bony defects in a LAP patient which when treated with a rather new bone graft material, along with PRF membrane yielded regenerative results on par with other graft materials. Nevertheless, the presence of very few local contributing factors should not be denied, though did not actually alter the significance of follow-up healing.

Conclusion
A clinically significant reduction in probing pocket depth with radiologic bone fill on either side with patient satisfaction was achieved. However, long-term clinical trials involving histological analysis of multiple cases to determine the efficacy of equine-derived bone graft would substantiate this case.

Declaration of Patient Consent
Informed consent was obtained from the patient and was also informed of all the photographs procured during the case study.

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Conflicts of Interest
There are no conflicts of interest.

References