



Comparative clinical and radiographic evaluation of autogenous bone grafts and demineralized freeze dried bone xenograft (Osseograft) combined with PRP in the treatment of human periodontal intra- osseous defects – 6 months study

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Abstract

Aim and objectives: The aim of the present study was to compare autogenous bone graft (Group - 1) and xenograft combined with PRP (Group - 2). The clinical and radiographic healing was evaluated after a period of six months in periodontal intra bony defects.

Material and methods: A split mouth design was followed, where two sites in the contra lateral quadrants with probing pocket depth of ≥ 5 mm with radiographic evidence of bone loss at baseline were chosen. Patients were grouped as **Group - 1:** 7 intra bony defects treated with autogenous



bone grafts, **Group - 2:** 7 intra bony defects treated with xenogenic grafts and platelet rich plasma (PRP).

Results: The mean plaque index score, oral hygiene index score, gingival index score showed statistically significant difference from baseline to 6 months. On comparison between Group - 1 and Group - 2 from baseline to 3rd and 6th month, mean PPD, CAL, defect fill was statistically highly significant with p-value <0.001. The percentage of mean PPD, CAL, defect fill was also assessed.

Conclusion: The surgical reconstructive treatment of intra-osseous defects with autogenous bone graft (Group - 1) resulted in clinically and statistically significant higher probing pocket depth reduction, clinical attachment level gain and radiographic bone fill compared to xenograft mixed with PRP (Group - 2).

Key words

Regeneration, Autografts, Platelet rich plasma (PRP), Xenografts.

Introduction

Periodontitis is an inflammatory disease of supporting tissues of the teeth caused by specific microorganisms resulting in the progressive destruction of periodontal ligament, alveolar bone with pocket formation, recession or both [1]. Conventional periodontal treatments such as scaling and root planing are highly effective in repairing disease related defects but result in the development of long junctional epithelium between the root surface and gingival connective tissue rather than regrowth of tissue that restores the architecture and function. Open flap debridement may result in the formation of long junctional epithelium which is more susceptible to microbial invasion and is thought to be less stable attachment. Thus bone grafting is the most common form of regenerative therapy [2].

Among all these biomaterials autogenous bone grafts have been adopted as gold standard since there is possibility to retain cell viability, graft revascularization and no possibility of disease transmission and contain live osteoblasts and osteoprogenitor cells and heal by Osteogenesis [3]. Autogenous bone grafts can be harvested either from intraoral or extra oral donor sites. Multiple intraoral locations have been used

including the maxillary tuberosity, exostoses, extraction sites and edentulous ridges [4]. Another important source of intraoral autogenous bone grafts includes the harvesting of osseous coagulum, bone blend generated from osteoplasty or ostectomy. The osseous coagulum is obtained by mixing the bone dust and blood [5]. This produces small particle size which induces more bone formation and provides additional surface area for the interaction of vascular and cellular elements. Recently, the autogenous bone scraper which is used in periodontics produces osseous coagulum of thin curled bone strips.

Xenograft is prepared by protein extraction of bovine bone that results in trabecular structure of hydroxy apatite similar to human cancellous bone. Allografts, Xenografts, Alloplasts do not possess inherent osteogenic properties and act only as a substrate for cell migration and proliferation and this led to the application of various biologically active substances. Polypeptide growth factors are biologic mediators that regulate cellular events including cell proliferation, chemotaxis, differentiation and matrix synthesis via binding to specific surface receptors [6]. PRP is a highly concentrated form of autogenous platelets and



works via the degranulation, providing a rich and readily obtainable source of diverse group of growth factors. PRP is believed to result in early consolidation and graft mineralization to increase the rate of bone formation and also promote 15–30% increase in trabecular bone density.

Thus in the present study, a combination of PRP and xenograft was compared with autogenous bone graft with respect to clinical and radiographic healing after a period of six months in periodontal intra bony defects.

Material and methods

A randomized, split mouth, single evaluator; 6 months prospective clinical study was conducted to evaluate the clinical and radiographic parameters in periodontal intra bony defects using autografts and xenografts with PRP. Informed consent was obtained from the patient and the ethical clearance was obtained from the institutional ethical board. Patients were selected from Out Patient Department of Periodontics, J.K.K. Nattraja Dental College and Hospitals, Komarapalayam using the following selection criteria.

Inclusion criteria

- Age limit of 20-50 years of both sexes.
- Probing depth ≥ 5 mm as assessed by Williams graduated periodontal probe.
- Patients with a minimum of two contra lateral intra bony defects.
- Vital teeth.

Exclusion criteria

- Known systemic diseases, short and long term drug therapies.
- Drug allergies.
- Pregnant and lactating women.
- Teeth with traumatic occlusion.
- Smokers.

Study design

A split mouth design was followed, where two sites in the contra lateral quadrants with probing pocket depth of ≥ 5 mm with radiographic evidence of bone loss at baseline were chosen. Probing pocket depth standardization was done with acrylic stent in all the selected areas.

- **Group - 1:** 7 intra bony defects treated with autogenous bone grafts.
- **Group - 2:** 7 intra bony defects treated with xenogenic grafts and PRP.

The following variables were measured at baseline, 3 months and 6 months post surgery.

- Gingival index (Loe. H and Silness. P, 1963)
- Plaque index (Silness. P and Loe. H, 1964)
- Oral hygiene index (simplified) (Green and Vermillion 1964)
- Probing pocket depth - deepest probing depth was measured.
- Clinical attachment level.

Radiographic parameters

An intraoral periapical radiograph (IOPA) of each defect site was exposed. Digitized images were displayed on the monitor at 5X magnification using Adobe Photoshop 7.0 computer software. A 0.5 mm grid was made on the digitized images and all linear measurements were made using Auto-CAD 2006 software.

Presurgical therapy

For all the enrolled patients routine blood investigations were taken. The initial therapy consisted of oral hygiene instructions, scaling and root planning. Three weeks following phase I therapy, a periodontal re-evaluation was performed.



PRP preparation

10 ml of blood was drawn from patients by a venipuncture of the antecubital vein 30 minutes prior to surgery. Blood was transferred to a sterile glass tube containing the anti-coagulant (3.8% sodium citrate). The initial centrifugation process was done at 1200 rpm for 15 minutes at room temperature. Serum was then pipetted out into new test tube and the remaining red cell fraction was centrifuged at 2000 rpm for 20 minutes. The second centrifugation resulted in

- Platelet poor plasma (PPP) on the top of the preparation which contains few platelets.
- Middle layer comprising of PRP which consists of platelets and White blood cells.
- Bottom most fractions comprising of red blood corpuscles which also contains newly synthesized platelet at the top most layer.

80% of PPP was pipetted out and discarded. PRP was then pipetted along with some red blood cell fraction and collected in a separate sterile glass tube.

Surgical procedures

The patient was anaesthetized using lignocaine 2% with 1: 1,00,000 epinephrine as a vasoconstrictor. Using Bard – Parker blade number 15 buccal and lingual sulcular incisions were made to elevate the mucoperiosteal flaps. Pocket epithelium and granulation tissue from the inner surface of flaps were carefully removed. Thorough soft tissue debridement and root planing were accomplished with Hu - Friedy curettes. The surgical area was then rinsed with copious amounts of sterile saline. With the autogenous bone scraper (Ebner grafter, Salvin Dental Specialists, USA) bone shavings were obtained from the site adjacent to the defect area. While scraping, shavings combined with blood and flow into collection chamber formed

osseous coagulum. Later the collected autogenous bone graft was grafted in the intra bony defect. (**Photo – 1 to Photo – 7**)

Photo - 1: Pre-operative view (Group - 1).



Photo - 2: Operative Procedure (Group - 1).



Photo - 3: Autogenous bone graft (Group - 1).



PRP was mixed with 10% calcium chloride to facilitate coagulation and to activate platelets before application. Osseograft (Advanced Biotech Products (P) Ltd India) is a demineralized bone matrix composed of type I collagen derived from bovine cortical bone samples (Xenograft), and particles of approximately 250 micrometers. Osseograft was emptied into a sterile dappen dish and PRP was added until the mixture becomes applicable. Increments of the graft material were added, to the bottom of the defect, and were condensed with an amalgam condenser to adapt the particles to the defect until it was completely filled. **(Photo - 8 to Photo - 14)**

After grafting, the flaps were approximated with simple interrupted sutures using 3-0 silk thread. Post surgical instructions were given to the patient and recalled after one week for suture removal and follow up.

Photo - 6: Pre-operative radiograph (Group - 1).

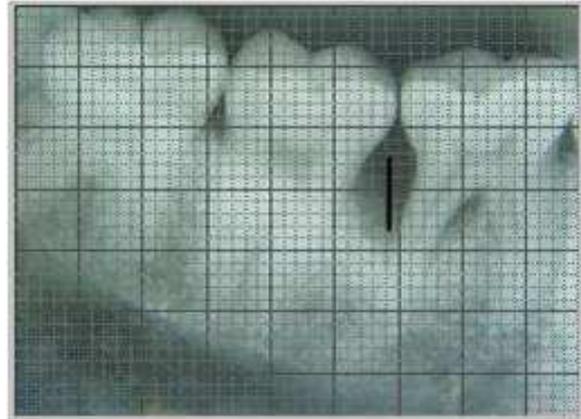


Photo - 4: Autogenous bone graft placed (Group - 1).



Photo - 7: Post-operative radiograph (6 months) (Group - 1).

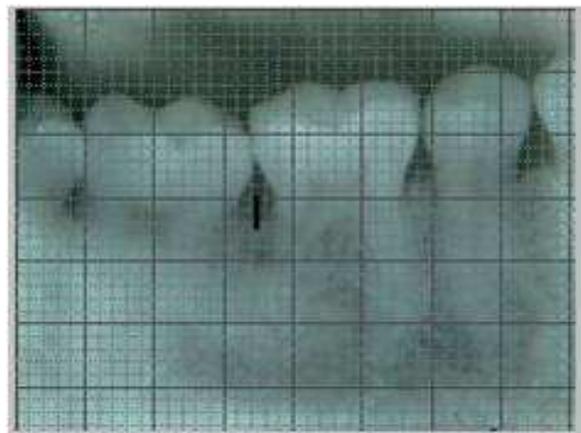


Photo - 5: Post operative (6 months) (Group - 1).



Photo - 8: Pre-operative view (Group - 2).



Statistical analysis

In this study Student t – distribution (William Sealy Gosset) was used to analyze the significance between the groups at different time intervals. $P < 0.001$ was considered as highly significant at 0.1% level of significance. $P > 0.05$ was considered as not significant at 5% level of significance.

index score was 1.02 ± 0.53 , reduced to 0.73 ± 0.48 , 0.42 ± 0.32 at the end of 3 months and 6 months respectively. The values at 3rd and 6th month were not statistically significant when compared to baseline; with a p-value > 0.05 as per **Table - 1** and **Graph - 1**.

Photo - 9: Operative procedure (Group - 2).



Photo - 11: Bone graft placed (Group - 2).

Photo - 10: Mixture of Xenograft and PRP (Group - 2).



Results

The mean plaque index score at baseline was 1.14 ± 0.68 , at 3rd month was 0.80 ± 0.52 and at 6th month was 0.51 ± 0.42 and the mean oral hygiene index score at baseline was 1.04 ± 0.70 , at 3rd month was 0.72 ± 0.52 and at 6th month was 0.52 ± 0.42 . At baseline the mean gingival

In Group - 1, at baseline the mean probing pocket depth was 7.85 ± 0.89 mm, reduced to 5.14 ± 0.75 mm at 3rd month and 3.86 ± 0.90 at 6th month. In Group - 2, at baseline it was 7.67 ± 0.80 mm, that reduced to 5.26 ± 1.20 mm at 3rd month and 4.57 ± 0.55 mm at 6th month as per **Table - 2** and **Graph - 2**. In Group - 1, the mean CAL at baseline was 8.71 ± 0.94 mm, at the end of 3rd month the gain was 6.14 ± 1.49 mm and at 6th

month was 4.57 ± 1.30 mm. In Group - 2, the mean CAL at baseline was 8.28 ± 0.60 mm, at the end of 3rd month was 6.83 ± 1.32 mm and at 6th month was 5.82 ± 1.12 mm as per **Table - 3** and **Graph - 3**. On comparison from baseline, the PPD reduction and the gain in CAL between the Groups at 3rd and 6th month time intervals were statistically highly significant with p-value < 0.001 .

Photo - 13: Pre-operative radiograph (Group - 2).

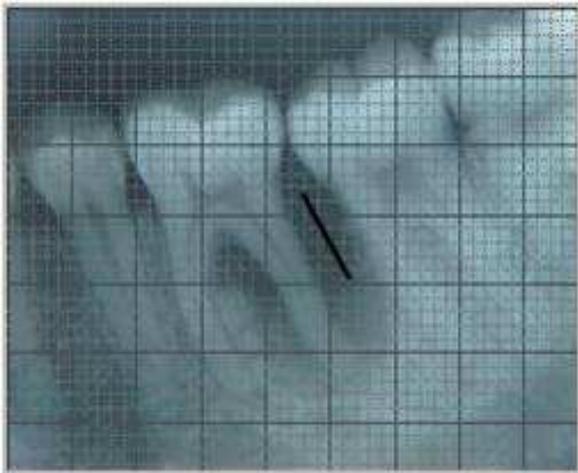


Photo - 14: Post-operative radiograph (6 months) (Group - 2).



In Group - 1, the defect at baseline was 7.57 ± 0.75 mm. The bone fill was 6.33 ± 1.11 mm and 4.42 ± 0.53 mm at 3rd and 6th month respectively. In Group - 2, the defect at baseline

was 7.71 ± 0.94 mm. The bone fill was 6.71 ± 0.75 mm and 5.57 ± 0.97 mm at 3rd and 6th month respectively as per **Table - 4** and **Graph - 4**. Comparing the mean defect fill between the Groups at 3rd and 6th month, statistically it was highly significant with p-value < 0.001 .

Discussion

The treatment of the periodontal diseases by traditional methods may result in healing by the formation of long junctional epithelium. Hence regenerative procedures have focused on the regeneration of new attachment apparatus including cementum, periodontal ligament and alveolar bone. In the present study autogenous bone graft was compared clinically and radiographically with demineralized freeze dried bone xenograft (Osseograft) combined with PRP in the treatment of human intra bony periodontal defects.

In the present study, at baseline, mean plaque index scores, oral hygiene index scores and gingival index scores were found to be 1.14 ± 0.68 , 1.04 ± 0.70 and 1.02 ± 0.53 respectively. All these values reduced to 0.51 ± 0.42 , 0.52 ± 0.42 and 0.42 ± 0.32 respectively at the end of 6 months. The changes at different time intervals (3rd and 6th months) was not statistically significant (p-value > 0.05). According to Gupta et al. (2007) [7], the patients undergoing periodontal therapy will maintain optimal oral hygiene and their compliance led to the improvement in their plaque index and gingival index scores.

In Group - 1, the PPD reduction at the end of 6 months was 50.82%. In Group - 2, it was 40.41%. In Group - 1, the gain at the end of 6 months was 47.53%. In Group - 2, at the end of 6 months was 34.54%. There was 12% increase in gain in Group - 1 compared to Group - 2. On comparison between the Groups at different



time intervals (3rd and 6th months), Group - 1 was statistically highly significant with p-value <0.001. It is believed that autogenous bone grafts produce pronounced revascularization and enhances osteogenesis which resulted in marked reduction in PPD and gain in the CAL compared to other bone graft materials [8, 9].

In the present study, Group - 1 resulted in 46.60% of bone fill at the end of 6th month. In Group - 2, the bone fill was 27.7% and at the end of 6th month. There was 19% increased bone fill in Group - 1 compared to Group - 2 which was statistically highly significant with p-value <0.001. This is in accordance with Becker et al. (1998) [10], who observed that the healing of demineralized bone grafts was delayed due to the retention of non-vital bone particles and was primarily osteoconductive in nature and only act as space filler particles in the defect site whereas autogenous bone grafts heals by osteogenesis and retains viable osteoblasts.

Particle size of bone grafts also plays an important role which results in greater bone fill, greater gain, and improved healing response in Group - 1. According to Coverly (1975) [7], the smaller and thinner particles have the advantage of making them readily susceptible to hydrolyzing enzymes which dissolve their cementing substances and liberate minerals. These particles also results in rapid resorption which can lead to a local rise in calcium concentration and thus favor more bone formation [11, 12]. In our study Group - 2 (xenograft and PRP) produced less PPD reduction, less CAL gain and decreased bone fill compared to Group - 1 (autogenous bone grafts). The results corroborate the findings of previous studies done by Schlegel et al. (2004) [13], who have shown that PRP increases bone formation only in the initial stages of healing. Nagata and Melo (2009) [14], also reported that after 4 weeks the direct influence of PRP will

fade away and physiological mechanisms of bone repair will continue to work according to the type of graft used.

Conclusion

The results presented here clearly indicates that surgical reconstructive treatment of intra-osseous defects with autogenous bone graft (Group - 1) resulted in clinically and statistically significant higher probing pocket depth reduction, clinical attachment level gain and radiographic bone fill compared to xenograft mixed with PRP (Group - 2). However, limitations of this study include a small sample size and limited time frame for post surgical evaluation. Hence it is necessary to have a large sample size and long term well controlled clinical trials to evaluate the true efficacy of these materials. Also further studies needed to be carried out to confirm the effects of PRP in periodontal regeneration.

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Table – 1: Comparison of mean plaque index scores, oral hygiene index scores, gingival index scores at baseline, 3 months and 6 months.

Parameters	Baseline	3 months	6 months	p - value
Plaque index	1.14 ±0.68	0.80±0.52	0.51±0.42	>0.05*
Oral hygiene index	1.04±0.70	0.72±0.52	0.52±0.42	>0.05*
Gingival index	1.02±0.53	0.73±0.48	0.42±0.32	>0.05*

* p- value between baseline, 3 months and 6 months was >0.05 denotes not statistically significant at 5% level.



Table – 2: Inter group difference in mean probing pocket depth (PPD) at baseline, 3 months and 6 months.

Probing pocket depth	Group - 1 (Mean±SD)	Group - 2 (Mean±SD)	p – value
Baseline	7.85 ±0.89	7.57 ±0.80	< 0.001**
3 months	5.14 ±0.75	4.86±1.20	< 0.001**
6 months	3.86±0.90	4.57±0.55	< 0.001**

Table – 3: Inter group difference in mean clinical attachment level (cal) at baseline, 3 months and 6 months.

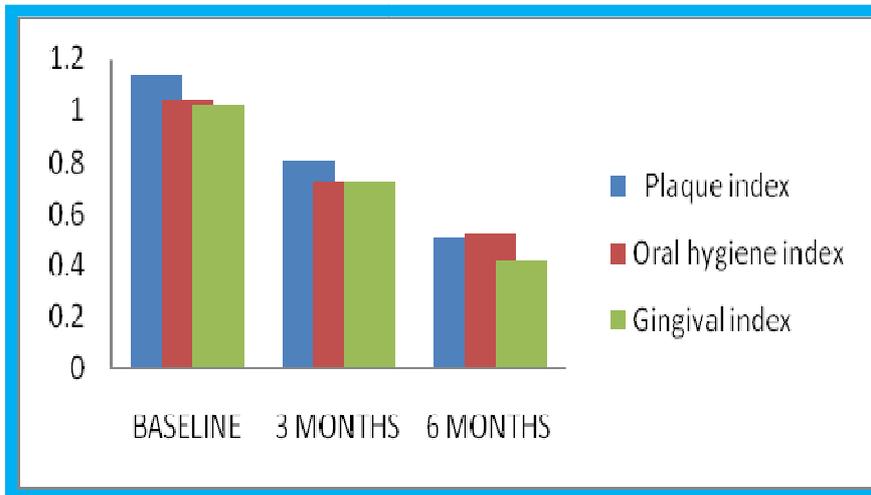
Clinical attachment level	Group - 1 (Mean±SD)	Group - 2 (Mean±SD)	p - value
Baseline	8.71 ±0.94	8.28 ±0.60	< 0.001**
3 months	6.14±1.49	6.83±1.32	< 0.001**
6 months	4.57±1.30	5.42±1.12	< 0.001**

Table – 4: Inter group difference in mean bone fill at baseline, 3 months and 6 months.

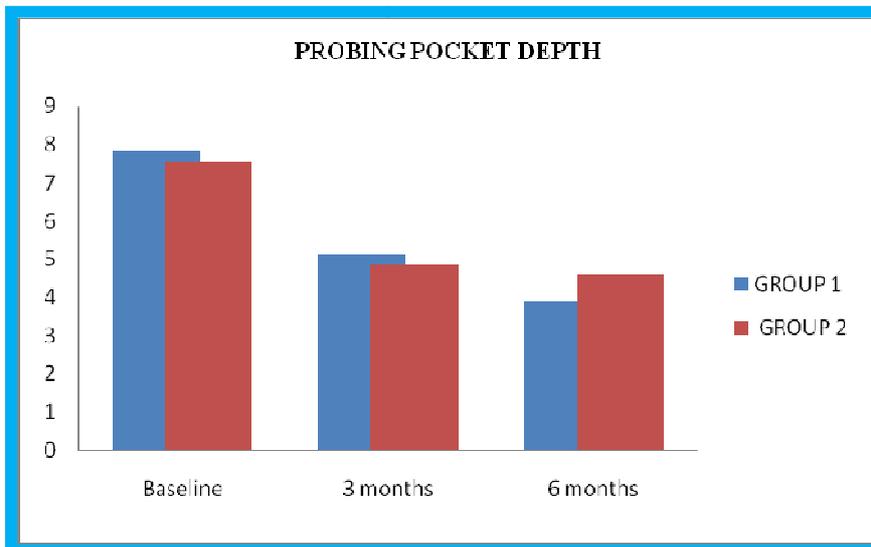
Defect	Group - 1 (Mean±SD)	Group - 2 (Mean±SD)	p value
Baseline	7.57 ±0.75	7.71 ±0.94	< 0.001**
3 months	6.33±1.11	6.71±0.75	< 0.001**
6 months	4.42±0.53	5.57±0.97	< 0.001**

** p- value between baseline, 3 months and 6 months was < 0.001 denotes statistically highly significant at 1% level.

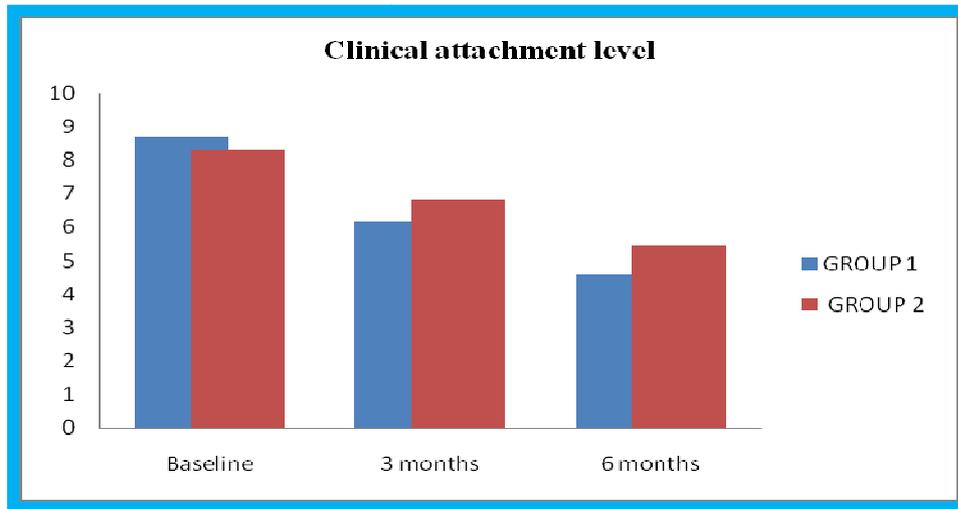
Graph - 1: Comparison of mean changes in plaque index, gingival index, Oral hygiene index at baseline, 3 months and 6 months.



Graph - 2: Comparison of mean changes in probing pocket depth (PPD) between the groups at baseline, 3 months and 6 months.



Graph - 3: Comparison of mean changes in clinical attachment level (CAL) between the groups at baseline, 3 months, and 6 months.



Graph - 4: Comparison of mean bone fill between the groups in baseline, 3 months and 6 months.

