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To Evaluate and Compare Clinically and Radiographically the Efficacy of an Alloplast (Biograft-HT®*), Platelet Rich Fibrin (PRF) and a Combination of Both in the Treatment of Intrabony Defects: An In Vivo Study

Lolakshi Kachroo[1*] Nisha[2] Aman Singh[3] Kanika Thakur[4] Tejinderjeet Kaur[5] Kriti Garg[6]

[1]Associate Dentist, Odontos Dental Clinic, Zirakpur, Punjab, India.
[2]Senior Lecturer, Department of Oral Medicine and Radiology, Rayat Bahra Dental College, Mohali, Punjab, India.
[3]PG Student, Department of Periodontics & Oral implantology, Faculty of Dental sciences, Desh Bhagat University, Mandi, Gobindgarh, India.
[4]PG Student, Department of Periodontics &Oral implantology, Himachal Institute of Dental Sciences, Paonta, HP, India.
[5]Private Practitioner, Ludhiana, Punjab, India.
[6]Senior Lecturer, Department of Conservative Dentistry and Endodontics, D.A.V Dental College, Yamunanagar, Haryana, India.

ABSTRACT

Background: To evaluate and compare clinically and radiographically the efficacy of an alloplast (Biograft-HT®*), Platelet rich fibrin (PRF) and a combination of both in the treatment of intrabony defects.

Materials and methods: A total number of 45 sites in 17 subjects were selected for the study and divided in three groups on basis of material use for treatment i.e platelet rich fibrin (PRF) and biograft-HT® and combination. Patients were allotted randomly to the above groups with the help of GraphPad software.

Results: The statistical analysis was done using SPSS (Statistical Package for Social Sciences) Version 15.0 statistical Analysis Software. The values were represented in Number (%) and Mean±SD. At baseline there was no statistically significant difference between the 3 groups, upon the studied parameters. This rule out any bias towards the outcome of the study.

Conclusion: Addition of platelet concentrate to the alloplastic material has no added benefit on bone levels when compared to the alloplastic and platelet concentrate alone.

Keywords: Bone graft, PRP, Bioplast , allograft, autograft.

INTRODUCTION

The purpose of the periodontal therapy broadly is to eliminate the inflammation of the periodontal tissues, arrest the destruction of soft tissue .[1] Bone grafting is one of the most common modality to restore the bony defects. Due to limitations of autogenous bone so, many attempts have been made to find a synthetic graft material that matches the features of autogenous bone.[2] Hydroxyapatite and tricalcium phosphate are the most widely used calcium phosphate biomaterials. Hydroxyapatite tends to favor the infiltration of bone and fusion to peripheral bone.[3]The rapid formation of new centripetal bone has been observed using porous hydroxyapatite.[4] **Biograft-HT®*** is a biphasic calcium phosphate ceramic consisting of hydroxyapatite and beta tricalcium phosphate in ratio of approx. 60:40 that is biocompatible, non toxic, resorbable, non inflammatory and bioactive.[5] PRP is prepared by complex techniques using hematology cell separators and two step centrifugation to concentrate platelets.

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***Correspondence Dr. Lolakshi Kachroo.**
Odontos Dental Clinic, Zirakpur, Punjab, India.
Email: lolakshikachroo888@gmail.com

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MATERIALS AND METHOD

A total number of 45 sites in 17 subjects were selected for the study from those attending the Out Patient Department of Periodontology and Oral Implantology. Complete medical and dental history of the patients, were obtained. Patients who were diagnosed as suffering from moderate to advanced periodontitis along with radiographic evidence of infrabony defect were selected for the study..

Inclusion Criteria:

1. Patients in the age group between 18 to 60 years, having moderate to advanced periodontitis.
2. Presence of at least one deep intraosseous defect with a clinical probing depth (PD) ≥ 5mm, with an osseous component of ≥ 3mm as measured radiographically.
3. Patients previously treated only by scaling and root planing and having good oral hygiene.

Exclusion Criteria:

1. Patients with a history of any systemic disease, interfering with the acceptance of the graft material and subsequent healing, or any other clinical contraindication for surgery.
2. Patients who had used antibiotics within 6 months prior to surgery.
3. Pregnant or lactating patients.
4. Patients who were smokers and were not willing to quit.
5. Patients with known allergy to drugs or materials used in the study.
6. Patients with platelet or any other bleeding disorder.

Methodology:

Patients included in the study were divided into three groups.

Group l: Defects treated with platelet rich fibrin (PRF)

Group ll: Defects treated with biograft-HT® (60% synthetic hydroxyapatite and 40% beta tricalcium phosphate). (IFGL Bioceramics Limited, Calcutta)

Group lll: Defects treated with a combination of PRF and biograft-HT®

Patients were allotted randomly to the above groups with the help of graphpad software.

Investigated Clinical Parameters

The following clinical parameters were recorded at baseline, 3 months post-operatively and 6 months post-operatively.

1. Plaque index **SILNESS AND LOE (1964)**

2. Gingival index **LOE AND SILNESS (1963)**

3. Pocket depth

4. Clinical attachment level

Probing measurements: pocket depth and clinical attachment level

Study models for all the selected patients were fabricated. A customized acrylic occlusal stent was prepared for each site to provide reproducible testing point and insertion axis. The stent was grooved in an occluso-apical direction to minimize variations in the direction of probing at subsequent recordings. The apical margin of the customized acrylic stent was used as the fixed reference point and the following measurements were recorded:-

1. Fixed Reference Point (FRP) to the Base of Pocket (BOP).

2. Fixed Reference point to the Gingival Margin (GM).

3. Fixed reference point to the Cemento-Enamel Junction (CEJ).

Reference point was used for better reproducibility. All clinical measurements were made by one examiner, using a manual calibrated UNC-15 periodontal probe.

The following calculations were made from the clinical measurements.

1. Pocket Depth = (FRP to BOP) - (FRP to GM)

2. Clinical attachment level = (FRP to BOP) – (FRP to CEJ)

Measurements were made pre-operatively at baseline and post-operatively at 3 months and 6 months, for all the sites.

Radiographic: Assessment and Measurements

The intraoral periapical radiographs of each defect site were taken pre-operatively and post-operatively using millimeter grid. Radiographic evaluations were done at baseline, 3 months and 6 months for the sites, as follows:

1. Distance from CEJ to the base of defect was measured - A (at baseline). A1 was taken at 3 months post-operatively and A2 was taken at 6 months post-operatively.

2. The distance from CEJ to alveolar crest was measured-B (at baseline). B1 was taken at 3 months post-operatively and B2 was taken at 6 months post-operatively.

3. Defect depth was calculated as C = A – B, C1 = A1 – B1 & C2 = A2 – B2 at baseline, 3 & 6 months respectively.

4. Amount of defect fill was calculated as X1= A - A1 at 3 months and X2 = A – A2 at 6 months, post-operatively.

5. Percentage defect fill was calculated as D1= (X1/A)x100 at 3 months and D2 = (X2/A) x100 at 6 months, post-operatively.

6. Amount of defect resolution was calculated as Y1= C-C1 and Y2=C-C2 at 3 and 6 months respectively.

7. Percentage of defect resolution was calculated as Z1=(Y1/C)x100 and Z2= (Y2/C)x100 at 3 and 6 months respectively.

Regenerative Materials Used in Study

Platelet rich fibrin

Platelet rich fibrin was obtained by centrifugation of venous blood. 8 ml of blood was drawn from a peripheral vein of the patient and collected into sterilized test tubes, without anticoagulant and the test tubes were centrifuged immediately at 3000 rpm (approx.) for 10 minutes. Resultant product consists of following three layers:

1. Top most layer is acellular platelet poor plasma (PPP).

2. PRF clot in middle.

3. RBCs at the bottom

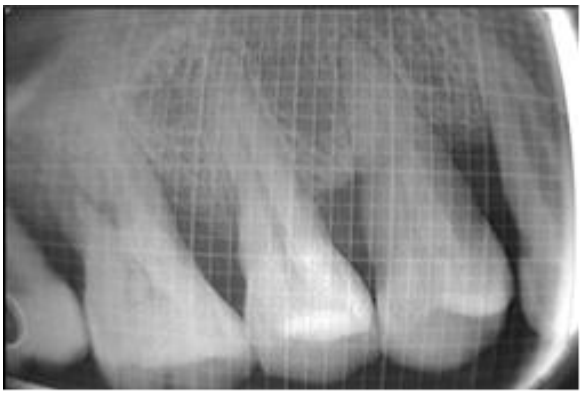

Fig 1: Post-operative radiographic picture of the defect at six months.

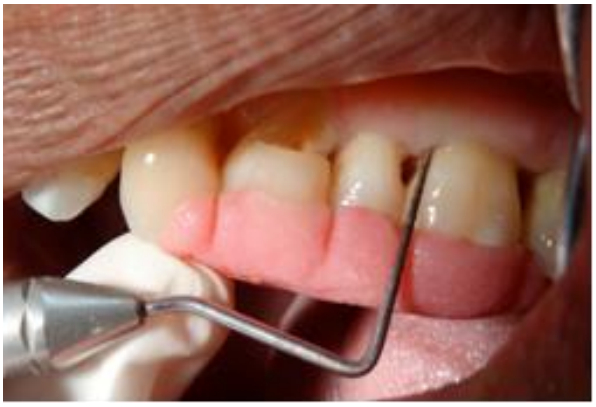

Fig 2: Post-operative clinical picture of the defect at six months.

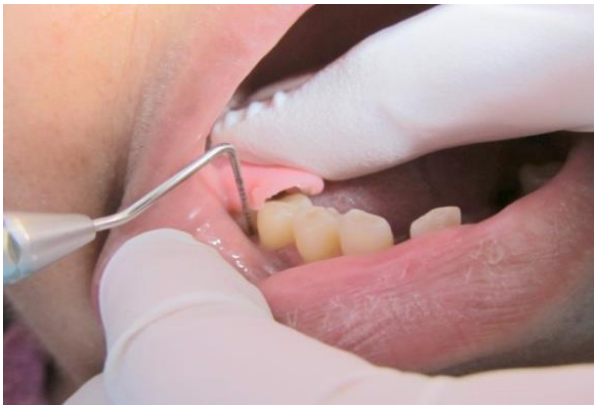

Fig 3: Post-operative assessments of clinical parameters at six months.

Preparation and Application of Graft Materials

For **group l** sites platelet rich Fibrin (PRF) was prepared. The middle layer of PRF was retrieved from the test tube and put in the dappen dish and then it was adapted to the defect site to reshape the

alveolar crest profile with the help of plastic filling instrument.

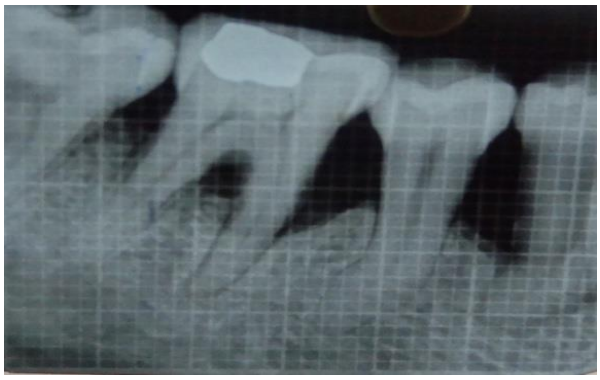

Fig 4: Post -operative radiographic picture of the defect at six months.

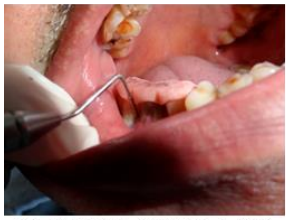

Fig.5: Post-operative assessment of clinical parameters at six months

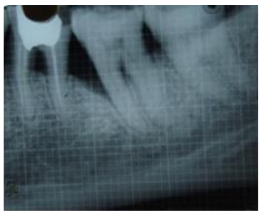

Fig.6: Post-operative radiographic picture of the defect at six months

In **group ll,** Biograft – HT® was placed into a sterile dappen dish and prepared by adding saline to obtain a paste like consistency. Then, small increments were added starting from the bottom of the defect and adapted well to its configuration. In **group lll** sites equal volume biograft-HT® and PRF were mixed as per the requirement of the defect, with the help of scoop. Then, small increments of this mixture were added starting from the bottom of the defect and adapted well to its configuration.

Pre suturing was done before placement of graft material. After graft placement, the soft tissue flap was repositioned at the original level and closed with interrupted direct loop sutures using 3-0 Mersilk sutures (Ethicon ltd.). Care was taken to achieve a tension free primary closure of flap while suturing. Surgical site was protected by covering it with periodontal dressing.

Post-Operative Management and Care

All the subjects were given both verbal and written instructions as a part of post-operative regimen.

1. The patients were advised to rinse with 0.2% chlorhexidine gluconate solution twice daily for 14 days for assistance in plaque control and to avoid chewing in the surgical area for the same period of time.

2. The patients were informed not to brush at the surgical site or manipulate it in any way for 10 days. They were prescribed medications which included non-steroidal anti-inflammatory agent ibuprofen 400mg thrice a day for post-operative discomfort and antibiotic Amoxicillin/clavulanic acid (625 mg thrice a day for 5 days) to prevent infection.

After 7 to 10 days dressing, sutures and any plaque present in the area were removed. The recall appointments were scheduled at 3 months and 6 months post surgically for soft tissue evaluation, plaque control, radiographic evaluation and for recording of clinical parameters.

RESULTS

The statistical analysis was done using SPSS (Statistical Package for Social Sciences) Version 15.0 statistical Analysis Software. The values were represented in Number (%) and Mean±SD.

Table 1a: Comparison of Clinical and Radiographic Parameters in the three groups under study at baseline was statistically non-significant (p>0.05).

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
PI	0.49	0.07	0.52	0.13	0.45	0.12	1.332	0.275	NS
GI	0.47	0.10	0.49	0.06	0.49	0.07	0.202	0.818	NS
PPD (mm)	5.53	0.64	5.47	0.83	5.40	0.91	0.103	0.902	NS
CAL (mm)	6.33	1.11	6.33	1.54	6.13	1.46	0.104	0.901	NS
Defect depth (mm)	6.93	2.22	5.67	1.72	6.33	1.59	1.737	0.188	NS

Table 2a: Comparison of PI scores in the three groups at different time intervals

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
Baseline	0.49	0.07	0.52	0.13	0.45	0.12	1.332	0.275	NS
3 months	0.54	0.06	0.54	0.14	0.48	0.13	1.526	0.229	NS
6 months	0.50	0.07	0.56	0.13	0.47	0.13	2.451	0.098	NS

Thus, the plaque score differences among the three groups were statistically non-significant at various time intervals.

In all the three groups, PI scores were reduced between baseline and 6 months. In Group II, all the comparisons showed a significant difference except for one between 3 months and 6 month interval.

Table 3a: Comparison of GI scores in three groups at different time intervals

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
Baseline	0.47	0.10	0.49	0.06	0.49	0.07	0.202	0.818	NS
3 months	0.56	0.06	0.55	0.11	0.53	0.14	0.277	0.760	NS
6 months	0.53	0.10	0.54	0.13	0.55	0.14	0.096	0.909	NS

Thus mean GI scores among groups at different time intervals were evenly matched. GI scores decreased from baseline to 3 and 6 months in all the three groups. The difference from 3 to 6 months for all groups was statistically non-significant.

Table 4a: Comparison of probing pocket depth in the three groups at different time intervals

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
Baseline	5.53	0.64	5.47	0.83	5.40	0.91	0.103	0.902	NS
3 months	3.13	0.92	3.27	1.16	2.73	1.03	1.064	0.354	NS
6 months	2.27	0.70	2.60	1.24	2.33	0.98	0.468	0.629	NS

Thus the mean probing pocket depth among groups at different time intervals was statistically non-significant.

Table 5a: Comparison of CAL measurements in the three groups at different time intervals

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
Baseline	6.33	1.11	6.33	1.54	6.13	1.46	0.104	0.901	NS
3 months	4.20	1.61	4.13	1.85	3.47	1.55	0.879	0.423	NS
6 months	3.33	1.72	3.53	1.51	3.07	1.62	0.314	0.732	NS

- Thus the difference in mean CAL measurements among the groups at different time intervals was non-significant.
- The comparison of CAL measurements between individual groups at different time intervals revealed statistically non-significant difference.

In Group I and Group II the change in CAL measurements was highly significant from 3 to 6 months but it was significant for Group III. In all the three groups, the change in CAL measurements was observed to be highly significant from baseline to 3 and 6 months.

Table 6a: Comparison of amount of defect fill in the three groups at different intervals

	Group I		Group II		Group III		ANOVA		
Parameter	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
3 months	1.80	1.21	1.40	0.51	1.27	0.59	1.677	0.199	NS
6 months	2.67	1.29	2.20	0.68	2.00	0.53	2.186	0.125	NS

Thus the difference in mean defect size was non-significant among groups at different time intervals. The Change in defect fill from 3 to 6 months was highly significant in Group II and significant ($p<0.05$) in Groups I and III.

Table 7a: Comparison of % defect fill in the three groups at 3- and 6-months interval.

	Group I		Group II		Group III		ANOVA		
Interval	Mean	SD	Mean	SD	Mean	SD	F	p	Sig.
3 months	27.09	16.09	26.39	11.39	21.35	11.15	0.861	0.430	NS
6 months	40.65	17.58	42.36	17.43	32.47	9.17	1.805	0.177	NS

Thus at both follow up intervals statistically there was non-significant difference in % defect fill among different groups. Difference was found to be non-significant statistically ($p=0.661$). Thus, the difference in mean defect size was non-significant among groups at different time intervals. For the defect resolution, between the group comparisons showed a statistically non-significant difference ($p>0.05$).

Thus at both follow up intervals statistically there was a non-significant difference in % defect fill among different groups. The percentage defect resolution between the individual groups was non-significant ($p>0.05$). In all the three groups % defect resolution increased between 3 to 6 months intervals, however, this increase was significant only in Groups I and II.

DISCUSSION

The combination of various regenerative biologic agents and techniques has recently attracted the interest of researchers in the field of reconstructive periodontal surgery.[6]PRF as an adjunct to allograft makes it possible to enhance the graft volume without injuring the maturation quality in new bone.[7] A total of 45 sites in 17 patients were selected in this study. Patients recruited in the study ranged between 21–59 years of age. Because of the slow rate of progression of chronic periodontitis, it usually becomes clinically significant in the third decade or later.[8]

The selection of patients with interproximal vertical defects was done using clinical and radiographic criteria similar to earlier studies of **Meffert et al (1985)[3], Kenney et al (1985)[1], and Shirakata M et al (2008)[9]**. The intraosseous defects ≥ 3 mm as observed radiographically and who had a probing depth of ≥ 5 mm

Mayfield et al (1996)[10] has observed better intra and inter individual reproducibility using a manual probe in comparison to simple and automated force-controlled probes (Florida Probe, Periprobe). **Zohar et al (2005)[10]** observed that surgical debridement of intraosseous defects lead to predictable increase in periodontal attachment of about 2.5 mm, with variable amounts of bone filling.

Nasr HF and Yukna RA (1999)[12] have stated that pre suturing reduces the possibility of displacing the bone graft during suturing process and also simplifies the procedure. Therefore, pre-suturing was done prior to placement of graft. Addition of graft into site was done in small increments to adapt the graft particles to the configuration of defect till the level of adjacent alveolar bone and condensed properly to ensure that the material reaches in the base of the defect and into the angles of the defect.

The soft tissue flap was repositioned at the original level and closed with 3-0 Mersilk suture using interrupted direct loop sutures to obtain primary soft tissue closure, as direct loop suture permits a better closure of interdental papilla.

The surgical site was covered with a non-eugenol (coe-pack) periodontal dressing for 7-10 days. According to **Sachs et al (1984)**[13] periodontal dressings protect the healing wound from coming in contact with food, saliva stagnation and trauma; thereby making post operative period comfortable. The patients were given both verbal and written instructions. Patients were prescribed amoxicillin with clavulanic acid 625mg postoperatively for 5 days.[14] Patients were given analgesic (ibuprofen 400mg thrice a day for 5 days), to avoid discomfort after the anaesthesia wears off.[15] Patients were instructed for ice application to prevent swelling. They were also instructed not to brush in the surgical area as it may lead to displacement of the periodontal dressing. Patients were advised to use 0.2% chorhexidine gluconate mouthwash twice daily for 14 days. According to **Addy M** [16] **and Newmann MG** [17] **et al,** chlorhexidine helps in reducing the bacterial load in the oral cavity and prevents plaque accumulation. The patients were scheduled for recall visits at 3 months and 6 months after surgery for the clinical and radiographical measurements.[18]

At baseline there was no statistical significant difference between the 3 groups, upon the studied parameters. This rules out any bias towards the outcome of the study.

Though in all groups, the scores improved from baseline to 3 and 6 month, this could was probably due to the good patient compliance and proper oral hygiene maintenance.

The results of the present study showed that all the graft materials resulted in increased bone levels, significant defect resolution, increased attachment level and reduced probing depth. The findings of study provide sufficient reasons to carry out more studies with longer time interval and larger sample sizes to confirm or reject the results of this study. Also more histologic studies are required to explore the biologic behaviour of this type of combination grafts.

CONCLUSION

Present study concluded that while the combination of the PRF and alloplast provided good soft tissue response with an additional effect on the CAL for intrabony defects. The results of the present study indicate that the addition of platelet concentrate to the alloplastic material has no added benefit on bone levels when compared to the alloplast and platelet concentrate alone. So, further, controlled, long term studies are needed to elucidate the role of adding platelet concentrates to graft materials.

CONFLICTS OF INTEREST

The authors declare they have no potential conflict of interests regarding this article.

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