



**Research Article**

**RIDGE PRESERVATION BY HYDROXY APATITE WITH COLLAGEN, PLATELET RICH FIBRIN AND CHORION MEMBRANE: A CLINICAL STUDY**

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**ABSTRACT**

**Objectives:** This study was primarily designed to determine the clinical efficiency of Hydroxy Apatite with Collagen (G-graft), PRF, Chorion membrane in preserving extraction sockets. To the best of my knowledge, there is not enough literature available that human chorion membrane was used for ridge preservation. **Materials and Methods:** For Control group (n=15) after debridement of the extraction socket, no additional treatment was performed. For Test group (n=15) after debridement, extraction sockets were filled with Hydroxyapatite with collagen mixed with PRF and was covered by a Chorion membrane and a cross mattress non absorbable suture was used to secure the membrane in place. **Results:** Clinically, there was a significantly greater decrease in the socket depth, whereas more decrease in buccolingual width of control than test group after 6months. There were statistically significant difference between extraction alone and ridge preservation groups for the Buccal, mesial and distal sites (P<0.05). The palatal/lingual site was relatively unchanged for either group, and there was no statistically significant difference in between groups (P>0.05). **Conclusion:** The use of G-Graft, PRF, Chorion membrane was effective in socket preservation. PRF being autologous, non immune, cost effective, easily procurable regenerative biomaterial. This study proved that chorion membrane was very effective as a barrier membrane.

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**INTRODUCTION**

Loss of alveolar bone may be attributed to a variety of factors, such as endodontic pathology, periodontitis, facial trauma and aggressive maneuvers during extractions. Most extractions are done with no regard for maintaining the alveolar ridge. Whether due to caries, trauma or advanced periodontal disease, tooth extraction and subsequent healing of the socket commonly result in osseous deformities of the alveolar ridge, including reduced height and reduced width of the residual ridge<sup>1</sup>. This bone loss appears to be progressive and irreversible. It is accelerated in the first 6 months leading to as much as 40% of the alveolar height and 60% of alveolar width loss followed by a gradual modeling and remodeling of the remaining bone.<sup>2</sup> In an attempt to preserve the alveolar bone and avoid the necessity of ridge augmentation prior to implant placement, various grafted materials have been used immediately following tooth extraction to fill and/or cover the socket. Some histological studies have reported positive healing responses with alloplast and xenograft, while others have shown negative results with demineralised freeze-dried bone allograft (DFDBA), bovine bone, and even autologous bone<sup>3</sup>.

Various authors<sup>4</sup> have described several surgical procedures ranging from regenerative techniques for socket preservation to immediate implant placement. Regenerative techniques have been widely tested in controlled and uncontrolled studies with various materials and clinical approaches: bone grafting alone, including autografts, allografts, xenografts, and alloplasts or in combination with absorbable or non-absorbable membranes. Recently, Surgiwear has developed xenograft in the name of G-Graft. It is natural Hydroxyapatite with natural collagen i.e bovine origin and with naturally occurring trabecular pattern. It is very useful for bone repair and replenishment. The shape can be changed by using Gigli saw and bone nibblers.<sup>5</sup> A recent innovation in dentistry is the preparation and use of platelet rich fibrin (PRF), a concentrated suspension of the growth factors found in platelets. These growth factors are involved in wound healing and are postulated as promoters of tissue engineering. PRF was shown to act as a suitable scaffold for proliferation of human periosteal cells in vitro, which may be suitable for applications in bone tissue engineering. PRF also induces the proliferation of various cells in vitro with the strongest induction effect on osteoblasts<sup>6</sup>. However, there is not enough literature available on the use of PRF in ridge preservation.

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One of the biomaterials used for scaffolds is fetal membrane. Amnion tissue is the innermost lining of the amniotic sac, the part of the placenta that encloses a fetus through term; chorion tissue is the next layer of the amniotic sac<sup>7</sup>. To the best of my knowledge, there is not enough literature available that human chorion membrane (Tissue Bank, Tata Memorial Hospital, Mumbai, India) was used for ridge preservation. To date, it is still uncertain as to which socket preservation technique is most predictable<sup>8</sup>. Collagen membrane in combination with allografts, xenografts and alloplasts has been used with varied success<sup>1</sup>. Recently, PRF because of its beneficial effects on healing has been used with different bone grafts. However, clinical data on the use of Hydroxyapatite with collagen, PRF, Chorion membrane for socket preservation is still lacking. In view of this, the present study was aimed at comparing the efficacy of Hydroxyapatite with collagen (G-GRAFT), PRF, Chorion membrane in post extraction sockets (Test Group) with extraction sites alone (Control Group).

## MATERIAL & METHODS

The present study was designed as a randomized, controlled clinical trial. The study sample was obtained from the Out Patient Department of Periodontics in G. Pulla Reddy Dental College & Hospital, Kurnool, India. A total of 40 patients (25 males, 15 females) who required tooth extraction in either the maxilla or mandible were selected based on the following inclusion and exclusion criteria:

### Inclusion Criteria

Male and female subjects of 18-50 years of age., extraction sites should have neighboring teeth on both sides, Grossly decayed teeth.

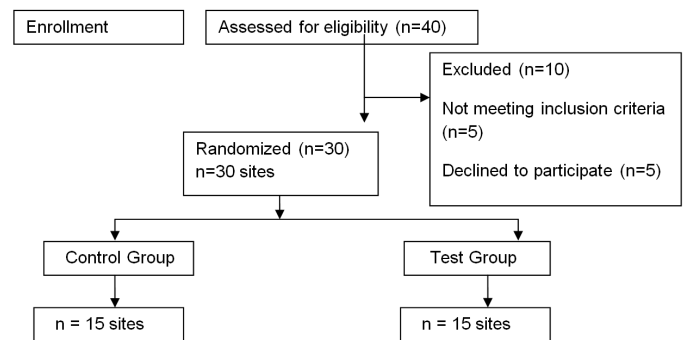
### Exclusion Criteria

- Systemic diseases including metabolic bone disease
- Patients with bleeding disorders.
- Patients with Aggressive periodontitis.
- Pregnant and lactating mothers.
- Patient on chemotherapy in the last twelve months or with history of radiation therapy.
- Current smokers or previous smokers.
- Long term steroidal or antibiotic therapy

### Study Protocol

After obtaining institutional ethical committee clearance the selected subjects were explained about the procedure and a written informed consent was obtained. Detailed medical and dental history of the patient was recorded. A total of 30 sites were treated. Only sites with adjacent teeth present were considered for the study. Study models and customized stents were prepared for each patient. The bone graft used in the present study was a sterile natural Hydroxyapatite with collagen (G-Graft).

## Study Design



The selected sites were randomly assigned utilizing a coin toss method into test & control groups. In the test group socket preservation was done using Hydroxyapatite with collagen (G-Graft), PRF and Chorion Membrane. In the control group extraction sites were allowed to heal by natural process.

### Preparation of Stent

On the study model the tooth to be extracted was marked. Self-cure clear acrylic was used by “sprinkle-on” method to prepare the stent. It extended one tooth mesially and one tooth distally. Buccally and lingually/palataly, the stent was limited to the occlusal one third only. Three grooves were marked in the stent corresponding to the midbuccal, mid lingual/palatal and mid occlusal point of the tooth to be extracted. The stents were preserved for every patient throughout the duration of the study by keeping it over the study cast.

### Clinical Parameters

- Following clinical parameters were assessed at baseline and at 6 months follow up.
- Plaque Index [Silness and Loe, 1964]
- Gingival Index [Loe and Silness, 1963]
- Horizontal width of the alveolar ridge was measured at base line as the distance between the buccal & lingual plate (buccolingually), using a bone calliper. At 6 months using occlusal stent as guide mark a point 2mm apical to buccal and lingual bone plate at mid socket region for measurements.
- Vertical ridge height was also measured at baseline & at 6 months. Measurements were obtained through markings made on the coronal portion of the stent at the mid portion of the socket, on the buccal plate & on the lingual plate using endodontic reamers with rubber stopper.

### PRF Preparation

Immediately prior to surgery, around 5 ml of whole venous blood is collected from the patients by venipuncturing of antecubital vein in each of the two sterile vacutainer tubes of 6 ml capacity without anticoagulant. The vacutainer tubes are placed in a centrifugal machine at 3000 revolutions per minute (rpm) for 10 minutes, Blood centrifugation immediately after collection allows the composition of a structured fibrin clot in the middle of the tube, just between the red corpuscles at the bottom and acellular plasma (Platelet Poor Plasma (PPP)) at the top. PRF clot thus formed was separated using sterile tweezers and scissors and transferred onto sterile gauze<sup>9</sup> (Figure 3).

**Initial Therapy**

Patients who met the inclusion criteria underwent phase I periodontal therapy that included scaling and root , oral hygiene instructions, temporary/ permanent restorations as required to enhance patients compliance, reduce the bacterial load, to prevent entrapment of calculus into the socket at the time of extraction.

**Surgical Procedure**

The surgical site was prepared with adequate anaesthesia using 2% lignocaine hydrochloride containing adrenaline in concentration of 1:80,000. A no 15 blade was used to initiate the preparation of the flap design by placing sulcular incision around the tooth to be extracted. The incision was extended one tooth mesially and one tooth distally. A full thickness flap was reflected using periosteal elevator. Periostomes were used to resect the periodontal fibers around the tooth. Extraction forceps were used to remove the tooth atraumatically (Figure 1& 2). Bone curette and Gracey curettes were used to debride the extraction socket. At this point base line measurements i.e. horizontal width (buccal to lingual bone margins) and vertical height of alveolar ridge(from occlusal stent to margins of buccal & lingual bone plate, mid portion of the socket) with bone calipers and endodontic reamers using acrylic stent as guide were measured.

**In the test group**

After debridement of the extraction socket, Hydroxyapatite with collagen mixed with PRF was inserted up to the level of the bone crest in the extraction socket and was covered by a Chorion membrane and a cross mattress non absorbable suture is to be used to secure the membrane in place. Sutures are removed at 7/ 10 days( Figure 4,5&6).

**In the control group**

After debridement of the extraction socket, no additional treatment was performed.

**Post Surgical Instructions**

Standard post-surgical instructions were given to the patient and they were instructed to rinse with 0.2% chlorhexidine digluconate twice daily for a 2- week period. Amoxicillin,500mg, 3 times daily was given for 7 days and analgesic medication ( Ibuprofen, 400mg) was prescribed. Patients were recalled 7/10 days after the surgery for suture removal and to evaluate wound healing. All the subjects were seen weekly until soft tissue closure over the site. All the parameters were evaluated again 6 months post operatively.

**Statistical Analysis**

All the parameters were recorded on a proforma/record sheet (appended). All the data recorded were subjected to statistical analysis using the SPSS software version 21. The results were averaged (mean ± standard deviation) for each parameter and are presented in tables. The following methods of statistical analysis have been used in the study. Comparison of mean values of plaque and gingival indices and other parameters among the two groups was done using t- test.

**RESULTS**

For this study a total of 40 patients were initially enrolled. 30 patients completed the study uneventfully. Maximum number

of patients were aged between 18 to 50 years. In Group I (Extraction alone) 42% of patients were male and remaining 58% were females whereas in Group II (G-Graft+PRF+Chorion Membrane)44% were Males and 55% were Females. The mean age of the patients in group I(Extraction alone) was 33.25±7.35 years as compared to34.56±7.36 years for patients in Group II( G-Graft+PRF+Chorion Membrane). Statistically, there was no significant difference between two groups. Thus the two groups were matched for age and gender. Papillae were preserved during the treatment procedures so that the mucoperiosteal flap covered the crestal alveolar bone. No attempt was made to cover the extraction socket or chorion membrane.

**Clinical Indexes**

The results of the mean plaque and gingival indexes are shown in Table 1. Both the groups showed a significant reduction in plaque and gingival indexes from baseline to 6 months. There were no statistically significant difference between the two groups.

**Horizontal Alveolar Ridge Width Changes and Vertical Ridge Height Changes**

Mean values for horizontal ridge width and vertical ridge height changes in preservation and extraction alone groups are shown in Table 2. Extraction alone cases had a mean initial alveolar width of 8.01±1.64 mm that reduced to 6.33±1.54mm at 6 months, for a statistically significant mean difference of 1.68mm (p< 0.05). Ridge Preservation cases had a mean initial alveolar width of 8.33±1.32mm that decreased to 7.36±1.10mm at 6 months, for a statistically significant mean difference of 0.97mm (p< 0.05). Mean between the groups approached a statistically significant difference(p<0.05).Mean and range for vertical changes in ridge height related to the original Buccal, palatal/lingual , mid portion of mesial and distal sockets. There were statistically significant difference between extraction alone and ridge preservation groups for the Buccal, mesial and distal sites (p<0.05).The palatal/lingual site was relatively unchanged for either group, and there was no statistically significant difference in between groups (P>0.05) (table 2).

**Histologic Evaluation**

Histomorphometric analysis of the trephine cores taken from the preservation sites at the time of implant placement showed predominantly cortical bone and minimal medullary spaces. Foci of amorphous, structureless, and eosinophilic deposits are evident, possibly remnants of graft material (Figure 7).

**Implant placement:** Surgical site was re-evaluated after 6 months followed by implant placement (Figure 8).

**Table 1** Summary of clinical Indexes over time by treatment group

Variable	Control Mean±SD	Test Mean±SD	P value
<b>Plaque Index</b>			
Base line	0.902±0.15	0.983±0.140	0.13
6 months	0.625±0.126	0.720±0.70	0.01
<b>Gingival Index</b>			
Base line	1.426±0.30	1.488±0.223	0.52
6 months	1.19±0.29	1.06±0.29	0.01

**Table 2** Summary of clinical parameters over time by treatment group

Variable	Control Mean±SD	Test Mean±SD	P value
<b>HRW</b>			
<b>BLW</b>			
Base line	8.01±1.64	8.33±1.32	0.123
6 months	6.33±1.54	7.36±1.10	0.001
<b>BBH</b>			
Base line	13.56±2.18	14.66±1.69	0.135
6 months	17.16±1.17	15.60±2.21	0.01
<b>LBH</b>			
Base line	13.36±1.82	14.63±1.73	0.06
6 months	14.80±1.77	14.73±2.07	0.71
<b>VRH</b>			
Mesial socket			
Base line	22.70±1.30	22.26±1.70	0.12
6 months	16.83±1.62	14.76±0.9	0.002
Distal socket			
Base line	21.46±1.87	22.4±1.18	0.114
6 months	16.06±1.36	14.53±1.18	0.0001

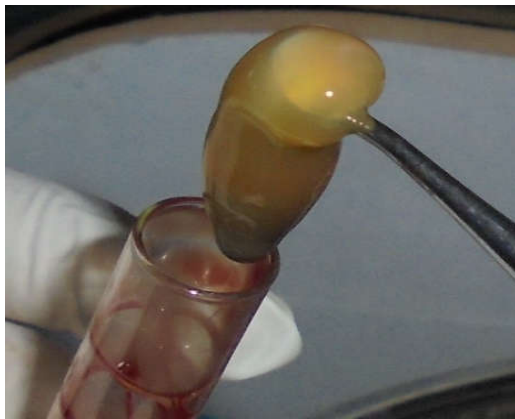
HRW= Horizontal Ridge width, BLW= Buccolingual width, BBH=buccoalbone plate height, LBH= lingual bone plate height, VRH= vertical Ridge height



**Figure 1** Tooth indicated for extraction



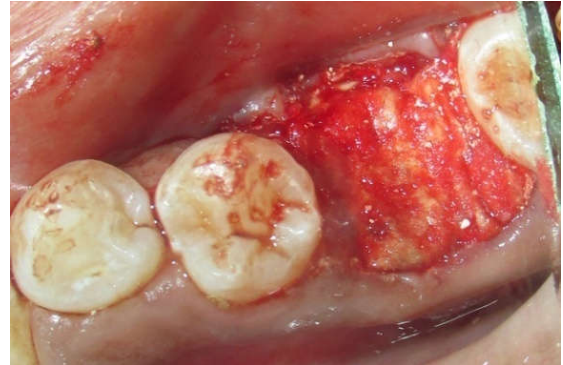
**Figure 2** After atraumatic extraction



**Figure 3** Platelet rich fibrin obtained



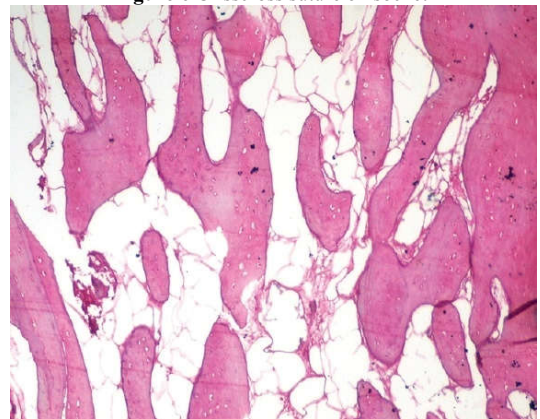
**Figure 4** G graft mixed with PRF in extraction socket



**Figure 5** chorine membrane placed over the socket



**Figure 6** Crisscross suture on socket



**Figure 7** Six month histologic image at x 10 for test group



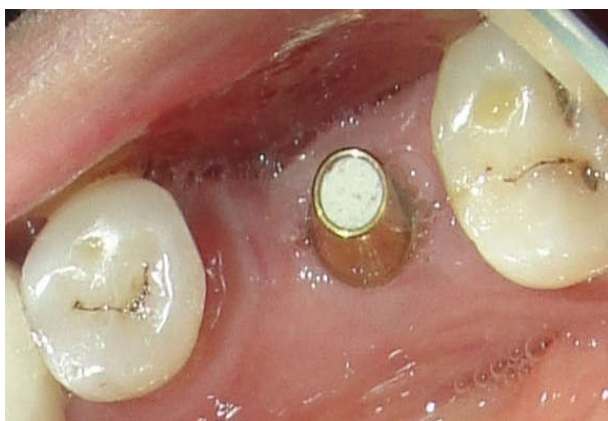


Figure 8 Implant placement after 6 months

## DISCUSSION

Alveolar ridge resorption has long been considered an unavoidable consequence of tooth extraction. There is a progressive loss of ridge contour as a result of physiologic bone remodeling. It is well accepted that as much as 40% of alveolar ridge height and 60% of alveolar width may be lost in the first 6 months following extraction.<sup>10</sup> According to Wolff's law Structural changes in the bone occur through cellular processes of osteoclastic resorption and osteoblastic deposition of collagen and subsequent mineralization of the collagen matrix. Ultimately, it is by modification of these mechanical, cellular, and molecular events that ridge preservation may be achieved following the loss of teeth.<sup>11</sup> Remodeling pattern causes a horizontal reabsorption that may also induce a further vertical reduction of the buccal bone.<sup>12,13</sup> To preserve alveolar bone and avoid the need for ridge augmentation, several materials were used immediately following tooth extraction to ensure the formation of alveolar bone within the sites. The ridge preservation procedure has been tested in controlled studies by Lekovic, Vance *et al*, Bin shi *et al*<sup>14,2,15,16</sup> with membrane alone or membrane plus graft, showing reduced ridge alteration compared to extraction alone. The present study was conducted to determine the efficacy of Hydroxyapatite with collagen (G-GRAFT), PRF, Chorion membrane in post extraction sockets (Test Group) with extraction sites alone (Control Group).

The mean age difference between the two groups was nonsignificant. In Group I (Extraction alone) 42% of patients were male and remaining 58% were females whereas in Group II (G-Graft+PRF+Chorion Membrane) 44% were Males and 55% were Females. Thus the two groups were matched for age and gender. Both the groups showed a significant reduction in plaque and gingival indexes from baseline to 6 months. There were no statistically significant difference between the two groups. Periodic recall visits during the study results in overall good oral hygiene in which patients were regularly reinforced for oral hygiene maintenance and underwent supragingival scaling if required. This is in accordance with the study done by Swati Das<sup>17</sup>, Jyostna Pinipe<sup>18</sup>.

In the present study after atraumatic extraction of the tooth base line measurements i.e. horizontal width (buccal to lingual bone margins) and vertical height of alveolar ridge (from occlusal stent to margins of buccal & lingual bone plate, mid portion of the socket) with bone calipers and endodontic reamers using acrylic stent as guide were measured. In the test group Ridge preservation procedure using Hydroxyapatite with

collagen (G-GRAFT), PRF, Chorion membrane were performed after complete debridement of the extraction socket. In the control group no additional treatment was done after debridement of extraction socket. Uneventful healing was observed with both the test and control sites with none of the patients reporting back with pain, swelling, dry socket or other complications.

Even though all efforts were taken to get maximum coverage of the membrane, complete coverage could not be attained with mucoperiosteal flap. The membrane was secured with several sutures. However, as shown in a study by Nam and Park in 2009<sup>19</sup>, membrane exposure during the healing period did not effect the efficacy of ridge preservation procedures.

This is the first study where chorion membrane was used as a barrier membrane in large sample size. A decrease in the buccolingual dimension of the alveolar ridge was observed in both control and Test groups. However it should be considered that, during the 6-month interval following tooth extraction, the alveolar ridge had undergone horizontal resorption that was significantly greater in the extraction alone groups than in the ridge preservation groups. The horizontal changes observed in this study were in agreement with Iasella<sup>20</sup>. The width of the Ridge preservation group decreased from  $8.33 \pm 1.32$  mm to  $7.36 \pm 1.10$  mm ( $P < 0.05$ ), while the width of the Extraction alone group decreased from  $8.01 \pm 1.64$  mm to  $6.33 \pm 1.54$  mm ( $P < 0.05$ ). Both the control and test groups lost ridge width, although an improved result was obtained in the RP group. These results shows that in the control groups i.e in Group A and Group B and in the test groups i.e in Group C and Group D there was a significant loss of buccolingual width more buccolingual width loss observed in control groups than test groups. The loss in vertical measurements was statistically significant in the control groups. There was also loss of vertical measurements in test groups but it was not statistically significant. The amount of socket depth fill was statistically significant in both test and control groups with a maximum of 7.9 mm in test group and 5.4 in the control group. When compared to the control group, test group had less dimensional change in the buccolingual width and the amount of socket depth fill was more in the test group.

Antonio Barone *et al* in 2008 observed similar postextraction alveolar ridge resorption in randomized, controlled clinical trial where extraction sockets were treated with either a porcine xenograft and a collagen barrier or freeze-dried bone and a collagen membrane and compared with the healing of 'empty' untreated extraction sockets<sup>21</sup>. More recent studies by Lekovic *et al*.<sup>2,14</sup> have shown that there is greater loss of alveolar ridge width than height and that some degree of loss was observed at all extraction sites.

The results of the present study were in accordance with the Van der Weijden *et al* where they conducted a systematic review to assess the amount of change in height and width of the residual ridge after tooth extraction. They found that the reduction in width of the alveolar ridges was 3.87 mm. The mean clinical mid-buccal height loss was 1.67 mm. The mean crestal height change as assessed on the radiographs was 1.53 mm.

In the present study, no effort was made to select a predetermined type of socket as in some previous studies<sup>22</sup>. The extraction sockets in this study presented with different soft tissue quantities, qualities and gingival tissue biotypes as well

as with different anatomical and dimensional characteristics of the hard tissue compartment. Obviously, some of these characteristics, together with several other factors may influence the final outcome of any socket preservation procedure and may be important in making the decision of whether or not a ridge preservation technique is indicated. The results of this study indicate that ridge preservation approach (using G-graft in combination with PRF and Chorion membrane) significantly limited the resorption of hard tissue ridge after tooth extraction compared to extraction alone.

## CONCLUSION

Regardless of the reasons for socket preservation, clinicians must be aware that sufficient alveolar bone volume and favorable architecture of the alveolar ridge are essential to achieve ideal functional and esthetic prosthetic reconstruction following implant therapy.

The results of this study revealed that ridge preservation procedure carried out immediately after extraction significantly reduced the alveolar bone resorption following tooth extraction when compared to extraction alone. The patient undergoing this procedure would be benefited by the presence of a ridge form that would allow for better esthetics, contour of fixed and removable prosthesis or even implant placement if necessary. The patient would also be spared the additional expense and trauma of ridge augmentation procedures to correct ridge defects.

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