EGYPTIAN DENTAL JOURNAL
www.eda-egypt.org
VOL. 70, 3183:3192, OCTOBER, 2024
PRINT ISSN 0070-9484 • ONLINE ISSN 2090-2360
ORAL SURGERY
Submit Date : 17-08-2024 • Accept Date : 17-09-2024 • Available online: 01-10-2024 • DOI : 10.21608/EDJ.2024.311258.3160

XENOGRAFT WITH ADVANCED PLATELET RICH FIBRIN COMPARED TO XENOGRAFT FOR MAXILLARY SINUS FLOOR AUGMENTATION: RANDOMIZED CLINICAL TRIAL

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ABSTRACT

Introduction: Sinus floor augmentation for posterior maxillary ridge augmentation to allow implant placement is a known procedure but with no exact consensus on the ideal grafting techniques. We hereby propose the use of A-PRF with Xenografts for sinus elevation.

Methodology: Eight patients were included in this study, randomly allocated into one of the groups; Control Group (Xenograft only) and a Study Group (a-PRF with the xenograft). The patients were prepared in the routine method and CBCTs ordered, and the study group patients and blood drawn and prepared according to the reported protocol to prepare a-PRF. Six months post-surgery, the patients were recalled CBCTs ordered and the implants placed with the elevated sinus floor , bone biopsies were also done at this point.

Results : The study showed comparable bone height after 6 months in both groups with that of the study group slightly higher but without a statistically significant difference. None of the patients reported signs of infection or perforation postoperatively.

Conclusions: Results of our study agree with reports proving that a-PRF promotes bone healing. Within the limitations of this trial; we hope to carry on our research in comparing the different platelet concentrates and their effect on bone regeneration. These may provide an autologous, cheap and easily prepared material to cover bone graft for ridge augmentation in maxillary sinuses or otherwise.

KEYWORDS: a-PRF, sinus augmentation, ridge augmentation, dental implants

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INTRODUCTION

Dental implants have now become the standard for treatment of edentulous spaces including those of the posterior maxillary region. Lack of sufficient bone needed for proper osseointegration of the dental implants has led to a myriad of research discussing methods to improve remaining ridge height, width or both [1,2]. Onlay grafting, sandwich osteotomies and sinus lifting/augmenting are only a few examples of these research propositions. Sinus augmentation is usually the treatment of choice in cases of deficient posterior ridge height and was first proposed by Tatum and has since led to the development of several editions to simplify and improve the initial surgical procedure [3]. The use of the lateral approach (historically known as the Caldwell Luc procedure) to augment the sinus is based on the access of the sinus membrane laterally, lifting it and either adding a grafting material or leaving it tented. The axial approach on the other hand is simpler but provides less exposure of the sinus membrane. The choice of approach is based on the ridge condition where a thinner/shorter ridge is an indication for a lateral approach sinus augmentation procedure.

The grafting materials that are used to augment the sinus include autogenous bone (from intraoral or extraoral sites), xenograft, allografts or a mix of these [4,5]. Autogenous bone being osteogenic, Osseo inductive and osteoconductive is the best choice but carries the increased risk of a second surgical site with its morbidity, bleeding time and longer surgical time [6,7]. Xenografts – especially those of bovine origin- are commonly used because of their biocompatibility and biological closeness to human bone. These are osteoconductive and provide excellent volumetric stability to the lifted membrane allowing for good bone fill for the second stage implant placement [8]. To improve the healing of the soft and the hard tissues; additives have been used to increase cellular proliferation, activation, migration, and differentiation. Growth factors such as Bone Morphogenic Proteins (BMPs) , Platelet derived growth factors (PDGFs), Transforming growth factor (TGFs)) and fibroblast growth factors (FGFs) are examples of these [9,10]. These are proven to improve bone and soft tissue healing at different surgical sites. PDGFs are found abundantly in platelet aggregates such as Platelet Rich Fibrin (PRF). PRF is an autologous fibrin adhesive with high platelet concentrations. Platelet rich plasma was the earliest reported form of blood-derived scaffolds with healing power. PRF was then developed with no cytotoxicity, higher cellular concentrate and a simpler preparation procedure. Choukroun's PRF reported in 2006 reportedly was acquired from human blood and it contained many types of blood cells (platelets, all types of WBCs, stem cells) in addition to growth factors [11]. Platelet concentrates have large amounts of essential nutrients, such as PDGF, TGFß2, TGFß1, VEGF. Also insulin-like growth factor (IGF), epidermal growth factor (EGF), and FGF which enhance cellular proliferation [12], matrix remodeling and angiogenesis were found in platelet concentrates. PRF is reported to enhance the formation of bone with grafts in sinus floor elevation / augmentation procedures [13] or even solely with sinus membrane tenting procedures. These cellular aggregates are produced from blood samples centrifuged at certain speeds and for specific time periods. It was later reported that lowering the centrifuging speed produces what was named A-PRF (advanced platelet rich fibrin) that has higher cellular concentration and better growth factor release than the previous PRF. [14-16]

We hereby hypothesize that the use of APRF with xenografts for maxillary sinus augmentation will provide better implant stability than xenografts alone.

METHODOLOGY

This study was planned as an RCT carried out on eight patients from the out-patient clinic of the Department of Oral and Maxillofacial Surgery. These patients were randomly allocated to two groups; the study group (Group I) to receive xenograft with A-PRF sinus augmentation while the control group (Group II) received xenograft augmentation. All surgical procedures and follow-up controls were performed between June 2021 and September 2022. The patients included in this study had bilateral missing posterior teeth with deficient alveolar ridges for implant placement. They were free from systemic and sinus diseases, non-smokers, above 18 years of age and with no parafunctional occlusal habits.

Presurgical preparation

The patients enrolled in this study were scheduled for a digital panoramic radiograph, complete blood analysis and primary alginate impressions for study cast fabrication. Written consents were obtained from all the included candidates and the local ethical committee approved this study 10-2022.

- ***Radiographic assessment***

Digital panoramic radiographs were ordered to assess the maxillary sinus and the remaining alveolar ridge height(crest of the ridge to the floor of the sinus perpendicularly)

- ***Blood analysis***

Complete blood count was requested to assure normal platelet count, red and white blood cell counts

- ***Study cast***

Alginate impressions were used to fabricate stone casts to assess occlusal relationships and interach space to plan implant placement.

First stage surgery

I. A-PRF preparation

Twenty milliliters of venous blood were drawn with a plastic syringe from the patient's non-dominant arm. The blood was gathered into two 10 ml dry glass tubes. The tubes; centrifuged at 1500 rpm, for14 minutes then obtain the A-PRF as reported in literature [*]. The A-PRF layer (which is the middle layer) is removed from the tubes and the cellular layer attached removed (Figure 1). The A-PRF from the first tube is cut and mixed with the xenograft while the second one is pressed between sterile glass slides to create a membrane. (Figure 2,3,4)

Fig. (1) After centrifugation; PPP, RBCs and PRF

* Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, Gogly B. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part I: technological concepts and evolution. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2006 Mar;101(3):e37-44. doi: 10.1016/j.tripleo.2005.07.008. Epub 2006 Jan 19. PMID: 16504849.

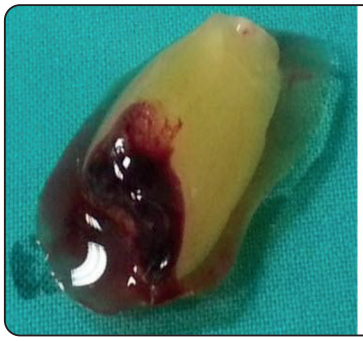


Fig. (2) Separated A-PRF

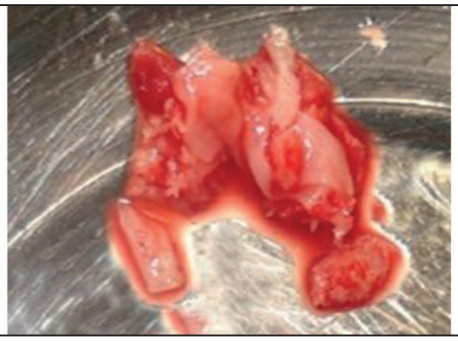


Fig. (3) Mixed with xenograft particles

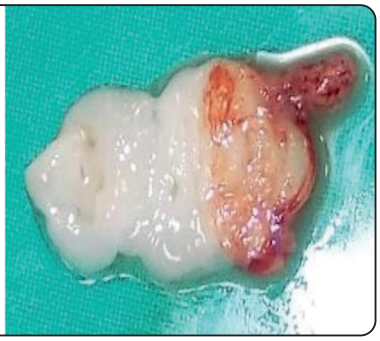


Fig. (4) Prepared A-PRF membrane

The patients used a Chlorhexidine Gluconate 0.1% mouthwash right before surgery. The surgeries were performed using local anesthetics (Articaine 4% with 1:100000 epinephrine) using a max. nerve block approach and buccal & palatal infiltrations. The maxillary sinus floor was accessed using a lateral approach with an anterior releasing incision. A bony window was made using a diamond stone bur under copious irrigation deep enough to expose the sinus lining membrane. (Figures 5,6).

The sinus lining was carefully stripped off the bone using broad curettes. The grafting materials were then placed in the cavity formed (A-PRF with xenograft or xenograft only in study and control groups respectively). The bony window was kept intact and then pushed inwards to create the new sinus floor at a higher level. In the study group the A-PRF membrane was used to cover the surgical site to keep the graft and A-PRF in place, while resorbable collagen sponges were used in the control group. The flaps were sutured back in place using vicryl 3-0 interrupted sutures. (figures 7-10).

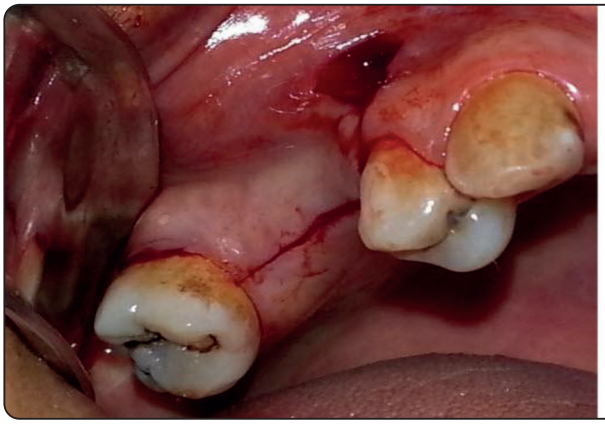


Fig. (5): crestal mucosal incision for access to the sinus floor

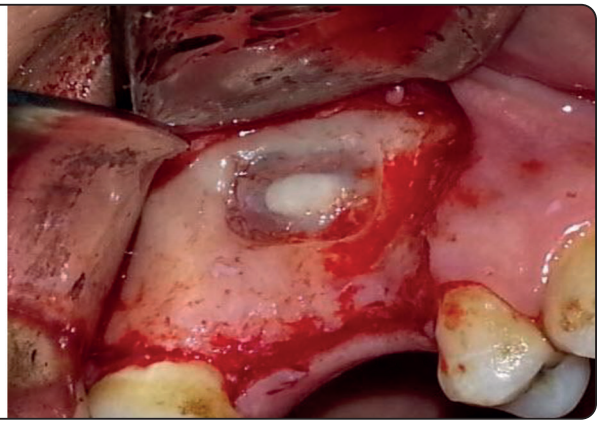


Fig. (6): exposure of the Schneiderian membrane

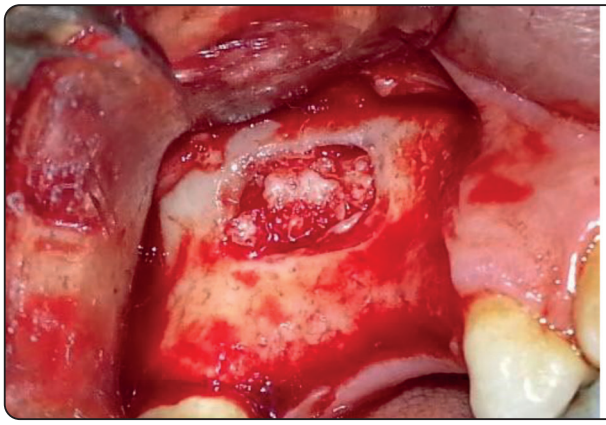


Fig. (7) Sinus filled with xenograft

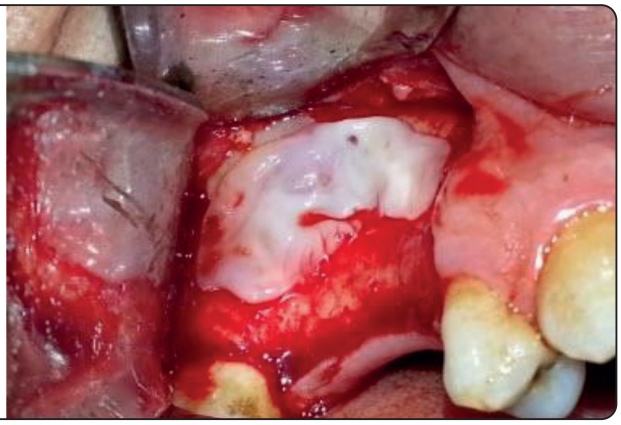


Fig. (8) the A-PRF membrane covering (Study side) .

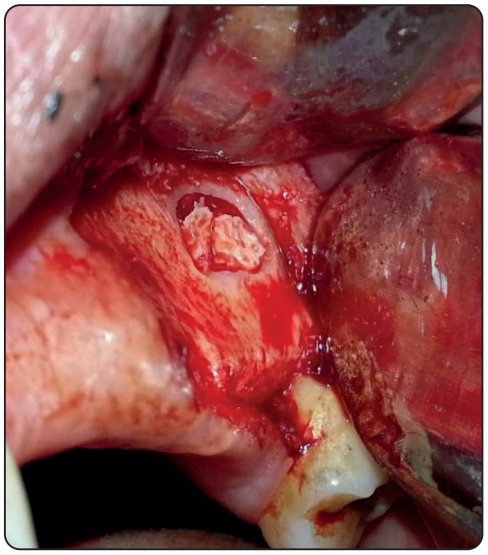


Fig. (9) Sinus filled with xenograft alone (Control side)

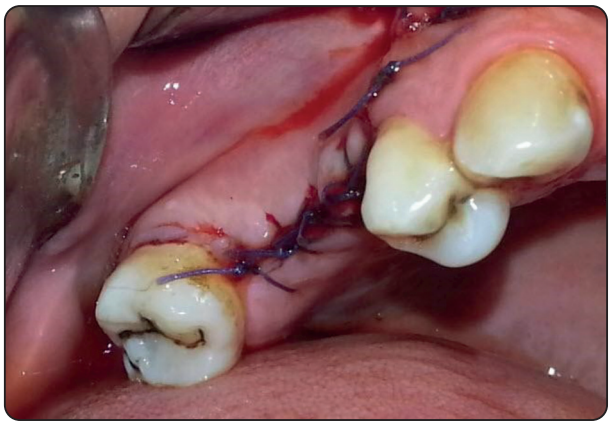
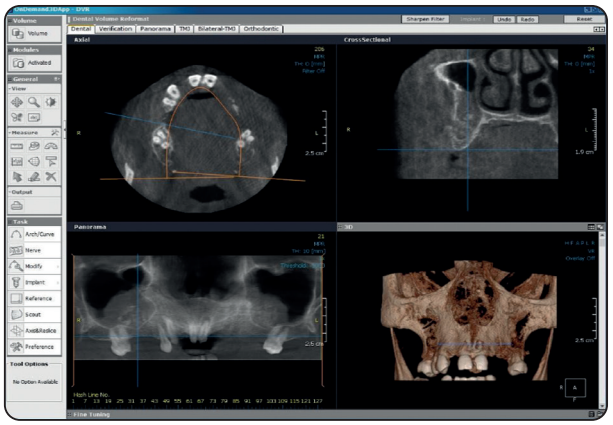


Fig. (10) Flap repositioning and suturing using vicryl 3-0 interrupted sutures

These patients were given postoperative instructions; to apply ice packs for the first 6 hours postoperatively. NSAIDS were prescribed 3 times daily for 4 days. Broad spectrum antibiotic was prescribes (3 times daily for 5 days). Oral hygiene measures and mouthwash were instructed, and the patients were told to avoid negative pressure such as using a straw or nose blowing.

The patients were recalled weekly then at 1,3,6 months post-surgery. Clinical assessment of surgical sites was done to ensure absence of inflammation and infection. CBCTs ant 1 week and 6 months were made and the bone height was re measured at these time points for comparison. (figure 11,12)



Fig, (11) Cone beam CT 1 week postoperatively

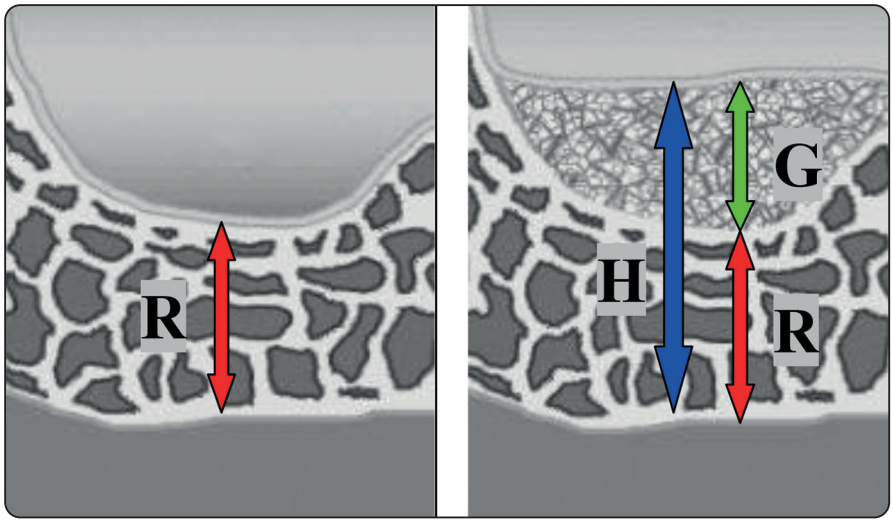


Fig. (12) Diagram showing radiographic measures. Residual bone height (R), Bone graft height (G), Total bone height (H)

Second stage surgery (6 months post-surgery)

The second stage was carried out 6 months after the initial 1st stage surgery. Biopsy harvesting and the placement of the dental implant were included in this stage*. (figure 13)

- Implant osteotomy was continued (at the sites where the biopsy had been taken)
- A ratchet is used to screw the implant until the intra-osseus part of it is totally inserted in the bone.
- Cover screw was then screwed in place in the fixture.

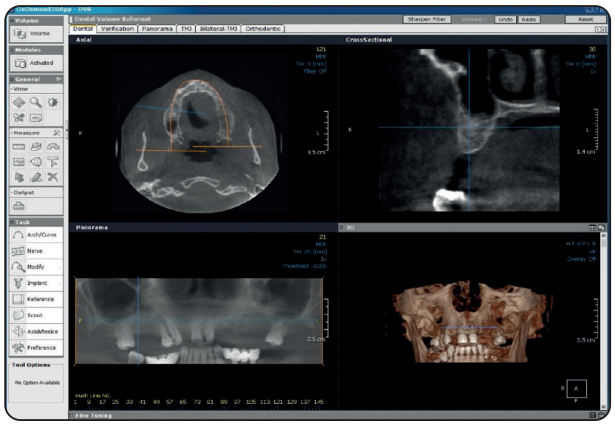


Fig. (13) Cone beam CT 6 months postoperatively

Biopsy harvesting

- A trephine bur of 3 mm diameter collected the transcortical biopsy out of the managed sinus.
- The trephine bur was introduced through the alveolar crest under copious amount of irrigation.
- The depth of the drill depth was derived from the preoperative CBCT to make sure that the biopsy included new and native bone.
- 10% buffered formalin was used to immediately fix the biopsy samples. (Fig 14)
- Implant placement
- The flap was finally sutured back into position with 3-0 vicryl.

Specimen processing

Decalcification of the specimen was done by suspension in EDTA 10% solution for up to 1 week. The solution was renewed daily. Once decalcified ; the biopsy was dehydrated with alcohol and cleared in xylol. Afterwards, it was placed in a paraffin to get a block. This was then sectioned vertically with a microtome, to thin paraffin slices (around 5 microns thick each). Masson Trichrome stain was used for histomorphometry analysis.

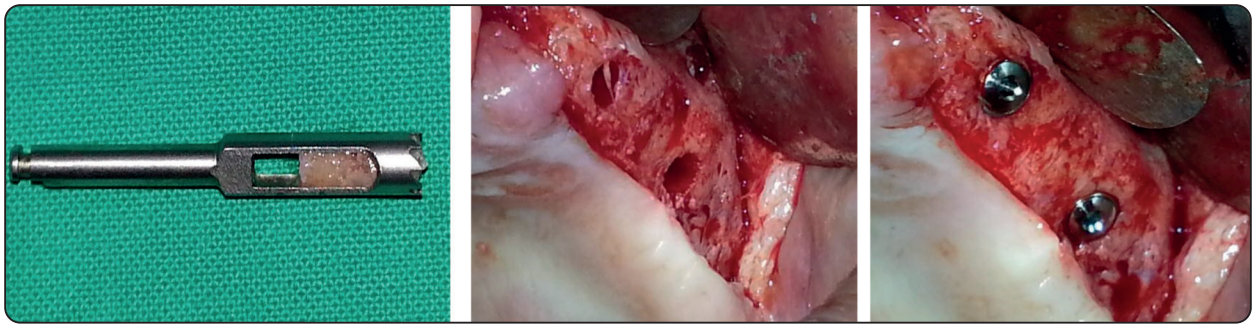


Fig. (14) (a) trephine bur with the bone core biopsy (b) implant osteotomy preparation (c) implant placement

Histomorphometric analysis

Newly formed bone and residues of the bone substitute as a % of the whole area was calculated at 40 power field. For the MT biopsies the new mineralized bone is green, and bone substitute residues red (Fig 15).

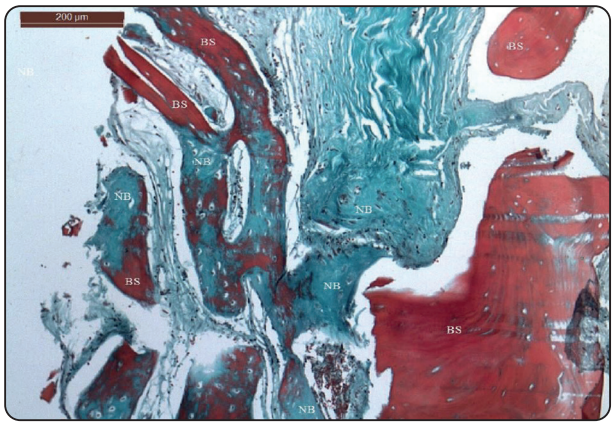


Fig. (15) Masson Trichrome stain: Mineralized trabecular bone appears green (NB), while xenograft residues appear red (BS).

Statistical analysis

Statistical assessment was done using SPSS. Data was provided as a mean and standard deviation. The student t-test was applied to compare different variables within the studied groups. Statistical insignificance was considered when P-value was > 0.05.

RESULTS

Our study was an RCT conducted on 8 patients (age 48.65 ± 9.36 years) with inadequate bone available for implant placement pointing out the need for maxillary sinus augmentation. A total of 16 sinus lift procedures were performed. The sinuses were allocated randomly into 2 groups. For intervention group, sinus floor augmentation was performed using A-PRF and xenograft, while for the control group augmentation was performed using xenograft alone. The surgical procedure was uneventful for all the patients with no obvious sinus membrane perforation (or except for 1 patient in the control group where small sinus membrane perforation occurred and was covered by collagen membrane). Healing period was uneventful for all the patients.

The intervention group showed slightly higher bone loss and bone loss percentage compared to the control group (3.46±1.4, 2.73±1.6 mm; 25.23±10.1, 21.13±14%), and there was no statistically significant difference between the two groups (P value 0.29, 0.46). (Table 1)

Newly formed bone was higher in the intervention group (19.5±7.6%) compared to the control group (15.4±7.2%). But no statistically significant difference was noted between the groups (P value 0.23). While both bone substitute and medullary space/connective tissue were higher in the control group (27.3±8.9, 57.3±10.6%) compared to the intervention group (23.8±5.3, 56.7±10.4%). But no statistically significant difference was noted between the two groups (P value 0.29, 0.89).

TABLE (1) Showing mean and standard deviation for bone height

	Intervention	Control	P-value
Residual bone height	4.45±1.89 mm	5.51±1.94 mm	--
Total height of bone after 1 week	18.19±0.92 mm	19.21±2.34 mm	0.29
Bone height of bone after 1 week	13.74±1.67 mm	13.7±3.42 mm	0.46
Total height of bone after 6 months	14.73±1.37 mm	16.48±2.33 mm	0.29
Bone height of bone after 6 months	10.28±1.85 mm	10.97±3.83 mm	0.89

DISCUSSION

The aim of this study was to assess the effect of addition of A-PRF to xenografts vs Xenografts alone without additives in sinus ridge augmentation. A-PRF is reported to have higher cellular content and better release of growth factors than other platelet derivatives [17]. The different derivatives (A-PRF ,L-PRF, P-PRF), are created using different centrifuge procedures carried out on the blood sample. Low speed centrifugation processes (15000 rpm for 14 minutes) provide what is now called A-PRF (Advanced Plasma Rich Fibrin) [18]. This is the protocol the authors followed in this study to acquire an A-PRF layer.

Xenografts are easy to use, require no second surgical procedure and so are advantageous in many cases. The major drawback is the risk of higher resorption rates. That is why the addition of growth enhancers is common practice. Results of this trial showed bone height gain in both control and intervention groups ; being slightly higher in the latter group but not statistically significant (14.73 mm and 16.48 mm respectively. This can be explained by the high content of IGF-1, PDGF, VEGF, FGF, EGF, platelet-derived epidermal growth factor and proteins of the fibrin matrix. These contents are proven to be of higher concentrates in A-PRF than other platelet concentrates [19]. Generally platelet concentrates were reported to induce probing depth reduction, and increase bone fill [17,20-22] .

In sinus floor elevation procedures with lateral approach, the need for acceptable bone regeneration for future implant placement is necessary. The use of PRF with sinus floor augmentation is well reported [23]. The use of a-PRF is yet to be tested widely to prove the better results and good bone regain enabling sinus floor elevation and implant placement[20,22,23]. With the easy preparation of a-PRF and the good results , our study agrees with such reports which proves good bone height and histopathologically adequate Haversian systems and mature bone was noted. Microscopically areas of bone formation (in the images above in blue) were seen with interspersed areas of red (remnants of the xenograft). This proves that the Xenograft + a-PRF combination successfully aided the elevation of the sinus floor.

Moreover when compared to the control group , where no additive was added to the xenograft; the total bone gain was statistically insignificant. We may explain this by the quality of the bone vs the quantity of the bone. The regenerated bone may prove to be of better quality in terms of maturity for example. Results of our study agree with reports proving that a-PRF promotes bone healing [17,21-23].

Within the limitations of this trial; we hope to carry on our research in comparing the different platelet concentrates and their effect on bone regeneration. These may provide an autologous, cheap and easily prepared material to cover bone graft for ridge augmentation in maxillary sinuses or otherwise.

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