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Effect of Platelet Rich Plasma (PRP) on Bone Graft in Alveolar Cleft Repair

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ABSTRACT

Background: Ultimate repair of alveolar defects is a great challenge in practical field, many bone graft materials have been evolved in the literatures for alveolar defect reconstruction as autogenous, allogenic, xenogenic, and alloplastic grafts **[1]**, autogenous iliac crest bone graft is the gold standard among other graft materials evaluated. However, the procedure associated with a potential risk of early complications such as graft resorption, graft leakage, infection, or graft failure, failure rate is about 30% **[2]**.

Objectives: To assess the efficacy of using Platelet Rich Plasma (PRP) in alveolar cleft reconstruction; in combination with Iliac Crest Bone Graft (ICBG) in comparison to the conventional Iliac Crest Bone Grafting (ICBG).

Patients and Methods: 20 patients underwent alveolar cleft reconstruction at the age of mixed dentition over a 3-year's period; their mean age was 8.8±2.3 years years and their mean post-operative follow-up was 13.4 months. Of these, 10 patients treated with ICBG combined with PRP (Group I), and 10 patients repaired by ICBG alone Group II (control group) results were assessed by rating the radiographs obtained 3, 6, and 12 months post-operatively according to cone beam CT (CBCT) volume and density assessment.

Results: Alveolar cleft repairs using PRP combined cancellous bone (Group I) were 90 percent successful, alveolar cleft repairs using cancellous bone (Group I) were 70 percent successful as regard; bone resorption reduction, bone volume gaining and improved bone density in CBCT at 3, 6, and 12 months post-operative with decreased post-operative complication rates.

Conclusion: Application of PRP enhances bone formation in alveolar clefts when admixed with autologous bone graft harvested from the iliac crest as it leads to early bone formation, increased bone density, decreases bone resorption, low infection rate and least post-operative complications.

Key Words: *Platelet rich plasma – PRP – Bone graft – Alveolar – Cleft repair.*

INTRODUCTION

Clefts of the lip, palate and alveolus are the commonest congenital anomaly to affect the orofacial region, for which efforts have been made to classify and repair, since the time of Veau 1931 **[3]**. Repair of cleft alveolus is an adjunctive procedure to further improve the functional and esthetic rehabilitation of patient with unilateral or bilateral cleft lip and palate, and is recommended during the mixed dentition period **[4]**. Clefting of the alveolar process is present in the majority of patients (75%) with cleft lip and palate. A perialveolar, oronasal fistula, commonly associated with the bony cleft, communicates between the alveolus, anterior hard palate, and floor of the nose **[5]**. Attempts for alveolar bone grafting go back as far as 1901 when Von Eiselberg used a pedicled bone graft to fill the defect. However, in 1908, Lexter was the first to demonstrate the use of free bone grafts. Various techniques for repair of the cleft alveolus have been described, including the use of bone graft **[6]**, periosteal pedicled flaps **[7]**, and free periosteal grafts **[8]**. The ideal age for alveolar reconstruction remains controversial; advocates of early procedures (including primary gingivoperiosteoplasty and primary bone grafting) as early cleft correction allows early orthodontic alignment of the teeth, correction of occlusion, and restoration of facial aesthetic balance without interference with subsequent maxillary growth arch results **[9]**. On the other hand early intervention causes later restrictions in maxillary growth, as elevation of the periosteum off the maxilla significantly retards growth of the maxilla **[10]**. The maxillary growth and dental age are the predominant considerations in determining the timing of alveolar reconstruction, maxillary growth is complete near the age of 8 years, whereas the maxillary canine does not usually erupt before the age of 10, though he agreed that grafting should be completed before canine eruption to allow for eruption of the tooth through the graft, thereby promoting stability of the tooth **[11]**. The American Cleft Palate-Craniofacial Association (ACPA) focused on treat-

ment of the alveolar cleft; (A) Early alveolar bone grafting in the first year of life with autogenous rib cortical graft as a separate operation **[12]**. (B) Presurgical Nasoalveolar Molding (NAM) with primary Gingivoperiosteoplasty (GPP) at the time of primary lip repair **[13]**; and (C) Secondary alveolar bone grafting as a separate operation during mixed dentition with autogenous iliac crest cancellous graft. (Standard) **[14]**. The concept of modern Presurgical Infant Orthopedics (PSIO) started with the work of McNeil, 1950 who was able to close the alveolar gap by using an acrylic appliance after taking maxillary impression **[15]**. Platelet Rich Plasma (PRP) is a new approach to tissue regeneration: It is widely used in various surgical fields, including head and neck surgery, otolaryngology, cardiovascular surgery, and maxillofacial surgery. Commonly, PRP is used in a gel form, which is formed by mixing PRP (derived from the centrifugation of autologous whole blood) with thrombin and calcium chloride. PRP gel includes a high concentration of platelets and a native concentration of fibrinogen **[16]**.

Cleft patients with unrepaired cleft alveolus suffer from nasolabial and palatal oronasal fistulae, collapsed alveolar segments, mobile premaxilla in bilateral cleft lip and palate, deficient bony support under the nasal ala, loss of periodontal support for permanent teeth adjacent to the cleft site, and missing, malpositioned, malformed, and supernumerary teeth **[17]**.

Repair of bony defects continues to be a challenging part of the many reconstructive procedures, although autogenously bone graft remains the standard in the reconstruction of bony defects, there are disadvantages including the limited amount of available bone and donor site morbidity, bone graft resorption and donor site morbidity, artificial bone substitute, as silicone, titanium phosphate may be used but expose the patient to the risk of foreign body reactions and infection **[18]**.

Our aim was to assess the efficacy of using Platelet Rich Plasma (PRP) in alveolar cleft reconstruction; whether added to the cancellous bone or not.

PATIENTS AND METHODS

After getting the approval from the Ethical Committee of Faculty of Medicine, Ain Shams University, twenty patients with alveolar clefts were collected randomly from the outpatient clinics of the Plastic Surgery Ain Shams and Minia University Hospitals from January 2014 to December 2018.

Their mean age was 8.8±3 years and their mean post-operative follow-up was 13.4 months, for each patient, age, sex, medical and surgical history was recorded.

All patients were examined pre and 3, 6 and 12 months post-operative by CBCT.

Only non syndromic patients between 6-13 years with unilateral ongenital alveolar defects were included, syndromatic and bilateral cases were excluded.

Patients divided into 2 equal groups; Group I included 10 patients treated by PRP added ICBG & Group II 10 patients treated by ICBG only.

In surgery incision was made around the labial surface of the fistula, first within the loose mucosa, then within the alveolar processes, and then continued along the margin of the alveolar cleft vertically toward the crest of the alveolus on each side.

Within the lesser segment, the incision extended to the second premolar, next the mucoperiosteum was dissected from the alveolar processes on the labial aspect using periosteal elevator to the nasal floor, exposing the lateral aspect of the anterior nasal spine and the lower pyriform rim, through the labial approach, the mucoperiosteum was elevated off the bony walls of the cleft, the oronasal fistula tract was then dissected and closed using interrupted 4/0 Vicryl sutures, then abox with mucoperiosteal linning is created to receive bone graft, then tension-free closure over the bone graft was confirmed.

The Cancellous Bone is harvested from the Iliac Crest using the standard technique, 5cm skin incision done 1cm posterior to anterior superior iliac spine, then incision advanced through the periosteum to expose the bone, and an osteotome was used to make a trap door fenestration (two vertical cuts and one from the medial aspect of the crest to connect the vertical cuts), then an anteriorly based cortical bone flap was elevated to expose the cancellous bone. A curette was then used to extract as much cancellous bone as needed. The cortical roof was then reduced back in place to cover the donor site and then closing the wound in layers by 4/0 vecryle.

Preparation of PRP: During the procedure 20cc of blood was withdrawn from the patient with Citrate Phosphate Dextrose (CPD) at a volume ratio of 9 to 1 for anticoagulation. PRP was pro-

duced through centrifugal separation of whole blood. After the first centrifugation (2500rpm, 5 minutes), the blood was separated into plasma and red blood cells. The red blood cells were discared, and after a further centrifugation (3500rpm, 10min) of the remaining plasma, the bottom layer, which is rich in platelets and constituted approximately 10% of the total withdrawn blood volume, was collected for use as PRP time of the solution was mixed with 2% calcium chloride at a volume ratio of 7 to 1 to activate platelets to release growth factors and 1cc of patient serum as a source of thrombin to achieve PRP in a gel form ready for use.

RESULTS

This study was performed on 20 patients with unilateral alveolar clefts; patients were divided into 2 groups; Group I (study group) included 10 patients had unilateral alveolar clefts treated with application of iliac crest cancellous bone graft mixed with PRP, and Group II treated by ICBG only. Patients had been evaluated clinically and by CBCT pre and 3, 6, and 12 months.

Pain and swelling (this pain and swelling only respond to strong non opiate analgesics, and anti-edema measures). Observed in 3 patients in Group I, and 6 patients in Group II. Also pain and swelling persisted longer in Group II than Group I. The difference was statistically significant *p*-value less than 0.05. Also discharge was present in two patients in Group I (20%) and 4 (40%) in Group II, but resolved within 7 days; the difference in both groups was statistically significant *p*-value less than 0.05, also time of resolution of pain and swelling is much longer in Group II than in Group I it takes about 2 weeks in Group II and about 10 days in Group I, after specific treatment as shown in Fig. (5).

In cone beam CT closure of the bony gap in Group II 60% of patients (six of ten patients) showed closure of the bony gap after 6 months and this percent raised to 70% after 1 year. In CBCT, in Group I (80%) of patients (eight of ten patients) showed closure of the bony gap after 6 months and this percent raised to 90% after 1 year. In CBCT with attachment of the two maxillary segments to achieve one piece of maxilla in spite of cleft one as shown in Fig. (3).

Quantitative evaluation of regenerated bone on CBCT and compared with controls, pre-operative, 3, 6, and 12 months post-operative, the average of the volume ratio of regenerated bone to alveolar cleft in cases with PRP was higher than in controls ($p<.05$), as shown in Figs. (4,5).

As regard eruption of canine tooth through the graft as indicator for the success of autogenous bone grafts was the eruption of the permanent canine adjacent to the cleft, which was considered erupted or nonerupted.

In this study Group II there were 5 erupted canine versus 5 nonerupted, in Group I there were 7 erupted versus 3 nonerupted the difference was significant $p<0.05$ as shown in Figs. (6,7).

The mean value for patient's pre-operative CBCT measuring (V pre) and post-operative CBCT examination (V post), to calculate difference value (Da=V post-V pre) and the relative difference value (Dr=V post/V pre X 100-100) (%). The mean Dr Value showed an increase by 93.73% on post-operative CBCT scans compared to pre-operative examinations in patients of Group I, and in Group II the mean Dr value was 76.26% as shown in Fig. (8).

Bone graft density at the cleft site rapidly increased till 6 months after the operation, and become stable at 12 months in both groups. But the graft denisty significantly increased in Group I more than Group II ($p<0.05$). The average bone density and bone graft height gain were 70% and 50% of normal at 12 months, in Group I and Group II respectively as shown in Figs. (9,10).

All over the study alveolar graft success as regard decreased resorption ratio in Group II patients treated by cancellous bone graft only were 70% successful, while in Group I patients treated by cancellous bone enhanced with PRP were 90% successful, with great significant statistical difference (*p*-value >0.05) as shown in (Table 1).

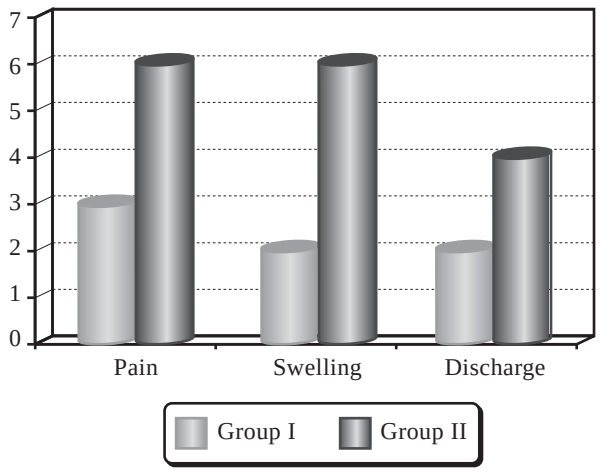

Fig. (1): Post-operative pain and swelling in both groups.

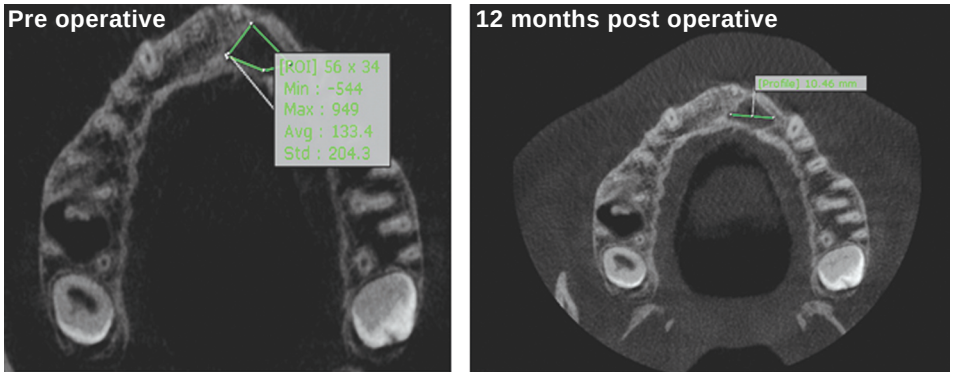

Fig. (2): Pre and 12 months post-operative CBCT of female with Lt side alveolar cleft treated by PRP added ICBG.

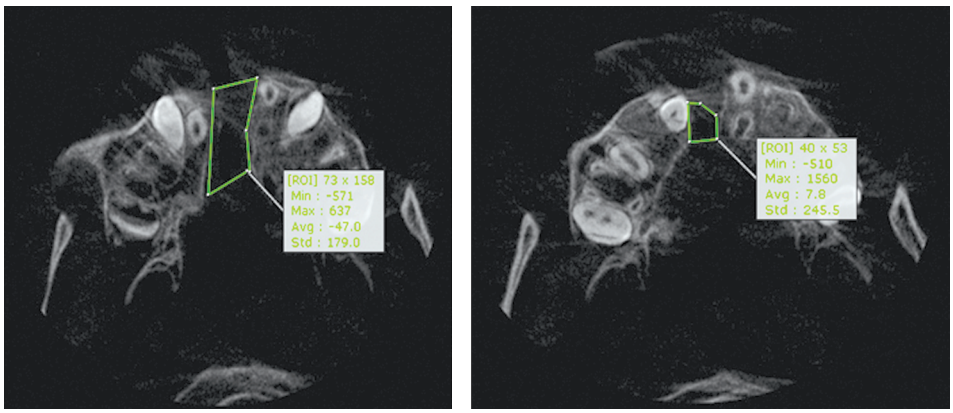

Fig. (3): Pre and 12 months post-operative volume assessment in CBCT in 11 years old male patient treated by ICBG combined with PRP.

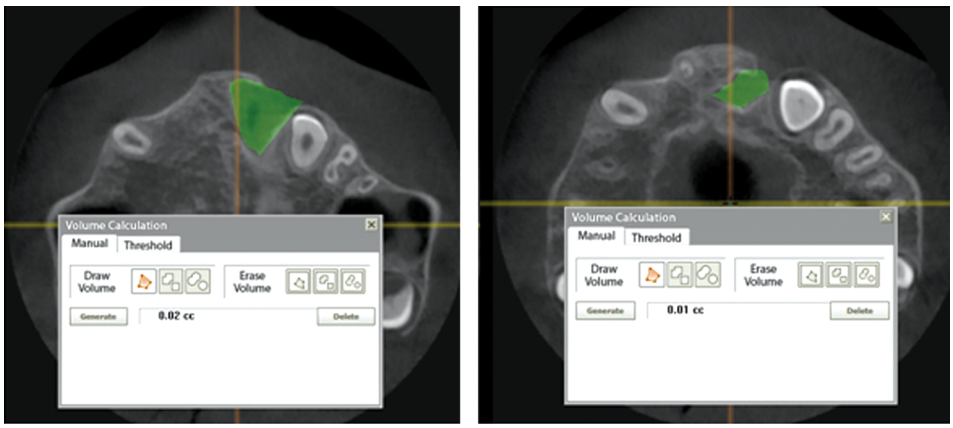

Fig. (4): CBCT pre and 6 months post-operative alveolar cleft volume in a 10 years old male with Lt side alveolar cleft repaired by ICBG and PRP.

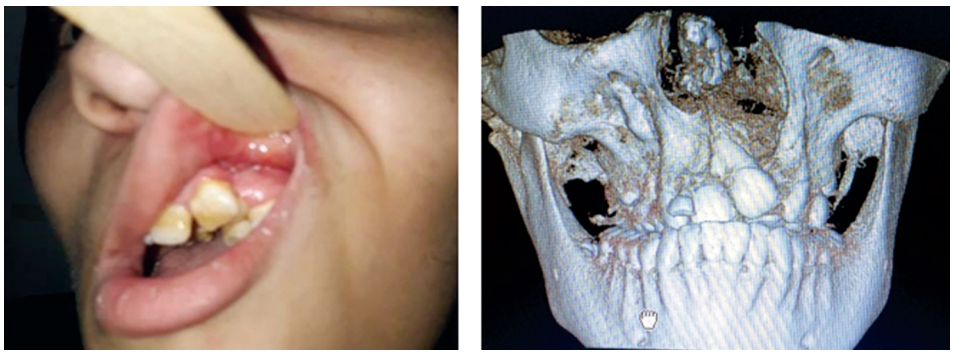

Fig. (5): Patient with persistent fistula in Group II treated with ICBG only after one year post-operative CBCT showing persistent alveolar defect.

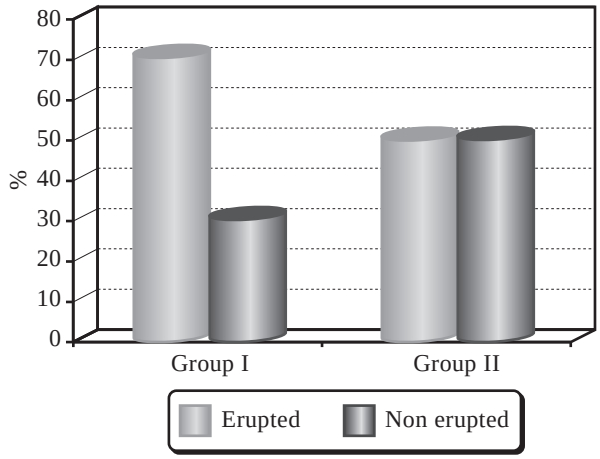

Fig. (6): Upper canine eruption at cleft site in both group.

Fig. (7): Post-operative CBCT film of 13 years old female from Group I with erupted Lt Canine.

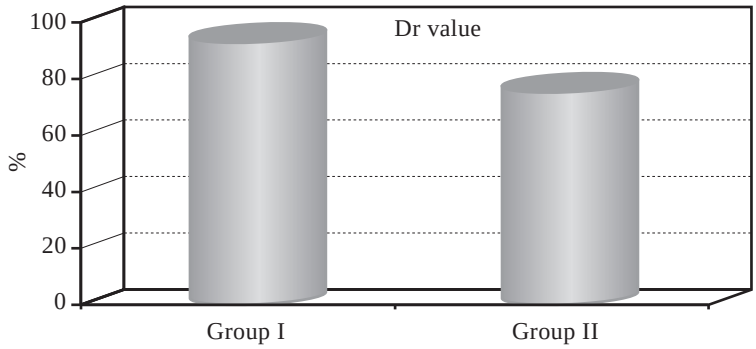

Fig. (8): Dr value (relative difference value of post-operative volume compared to pre-operative volume) in both groups.

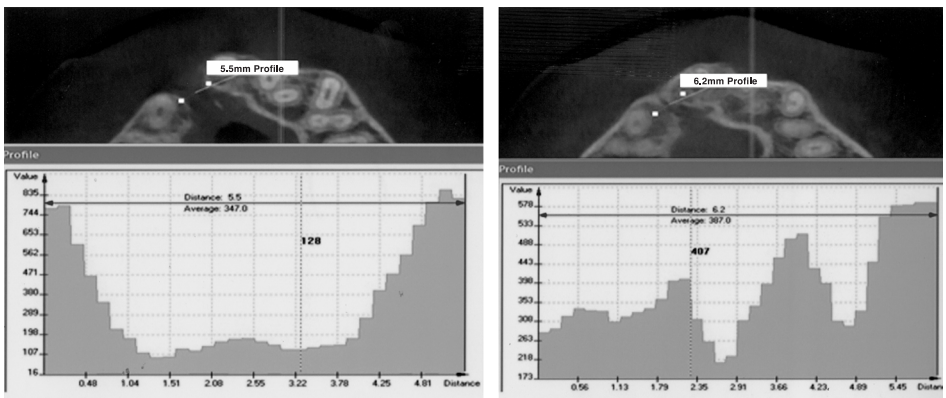

Fig. (9): Pre and 12 months post-operative bone density in CBCT for 12 years female patient with Rt side alveolar cleft repaired by ICBG combined with PRP.

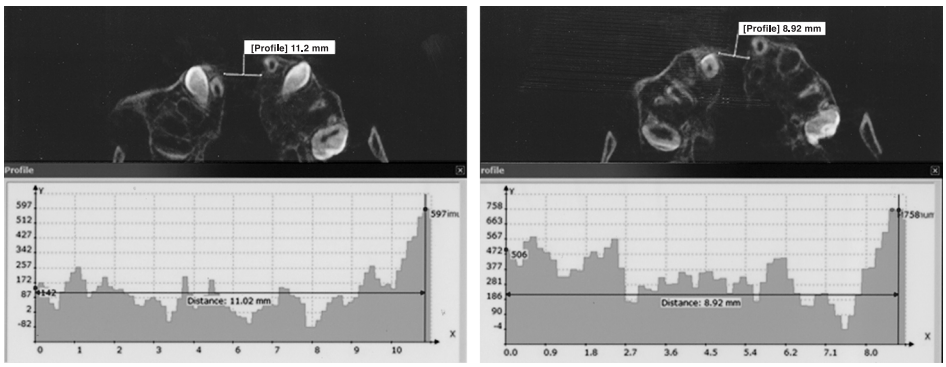

Fig. (10): Pre and 12 months bone density in CBCT for 10 years old male with Rt side alveolar cleft repaired by ICBG only.

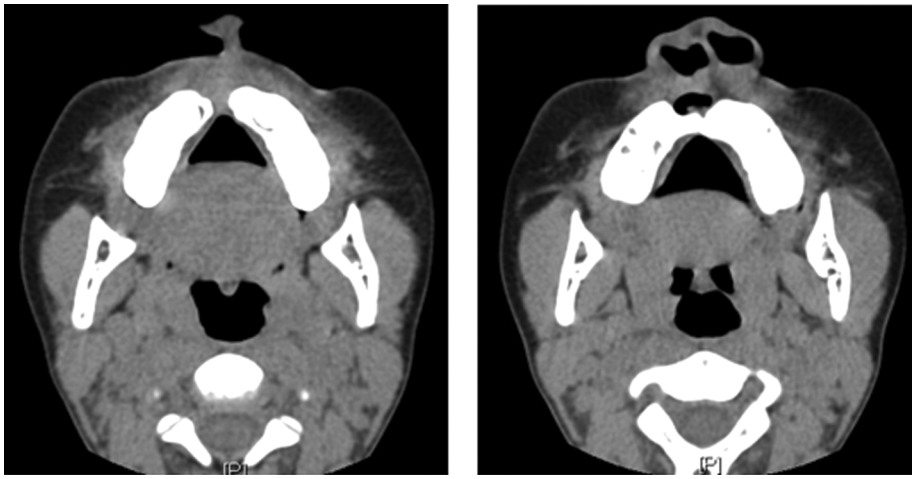

Fig. (11): Pre and 12 months post-operative CBCT of 11 years old male with Rt side alveolar cleft treated by PRP added ICBG.

Table (1): Resorption ratio at 3, 6, and 12 months in all study cases.

Case	V1 pre-op. vol. of cleft cm^3	V3; 3 months (RR) RR=Resorption Ratio	V6; 6 months (RR) RR=Resorption Ratio	PRP	Age in years
1	1.73	1.41 (18.5%)	1.31 (24.2%)	No	7
2	1.53	1.42 (7.2%)	1.33 (13.1%)	Yes	7
3	1.56	1.06 (32.1%)	0.99 (36.5%)	No	9
4	1.33	1.03 (22.6%)	0.93 (33.1%)	No	8
5	1.38	0.89 (35.5%)	0.72 (47.8%)	No	10
6	1.44	1.11 (22.9%)	1.02 (29.2%)	Yes	7
7	2.16	1.99 (7.9%)	1.75 (19%)	Yes	11
8	1.32	1.06 (19.7%)	0.89 (32.5%)	No	10
9	1.93	1.49 (22.8%)	1.19 (38.3%)	No	12
10	1.73	1.37 (20.8%)	1.22 (29.5%)	Yes	10
11	1.54	1.24 (19.5%)	1.06 (31.2%)	No	7
12	1.87	1.35 (27.8%)	1.13 (39.6%)	No	8
13	1.76	1.42 (19.3%)	1.29 (26.7%)	Yes	7
14	1.93	1.63 (15.5%)	1.55 (19.7%)	Yes	10
15	1.98	1.57 (20.7%)	1.43 (27.8%)	Yes	8
16	1.75	1.45 (17.2%)	1.15 (35%)	No	12
17	1.88	1.65 (13%)	1.38 (27%)	Yes	7
18	1.95	1.56 (20%)	1.33 (30%)	No	11
19	2.05	1.69 (17%)	1.59 (22%)	Yes	7
20	1.45	1.30 (10%)	1.13 (22%)	Yes	10
Mean	1.68±0.26	1.29±0.23 (21.78±6.88%)	1.15±0.23 (30.66±8.97%)		12

DISCUSSION

Secondary alveolar cleft bone grafting (SABG) has become a well-established treatment modality in the rehabilitation of patients with cleft lip and palate, closure of alveolar bone defects is necessary to allow orthodontics to restore a normal alveolar arch, allow dental and occlusal restoration in cleft and palate patients, and allow further orthognathic surgery **[19]**.

In the present study, 20 non syndromic cleft patients with unilateral alveolar cleft were divided into 2 groups. Group I were treated by closure of the defect with application of iliac crest cancellous bone graft combined with PRP, and Group II were treated by application of the iliac bone graft only.

In this study follow-up after 3, 6 months and 1 year was done by clinical evaluation, and Cone Beam CT to assess the closure of the gap with bone formation.

Discharge was seen in fewer patients when PRP was used because PRP primarily due to its acidic pH (6.5-6.7) inhibits bacterial colony growth. Secondarily, PRP concentrates WBCs and platelets to cause bacterial inhibition by greater number of

functionally viable leucocytes. Thirdly, rapid development of granulation tissue by early in-growth of capillaries prevents bacterial growth by bringing in circulating macrophages and neutrophils. Thus, creating an oxygen rich environment suppresses the growth of anaerobic micro-organisms.

In patients of Group II, infection occurred in 30% of patients, those patients received systemic antibiotics with antiseptic mouth wash but no loss of the grafted bone occurred with disruption of sutures. One patient had secondary closure later without need of another bone graft. All patients were closed successfully. In patients of Group I, infection occurred in 10% of patients and along with success of conservative management by systemic antibiotics and mouth wash for two weeks, post-operative bleeding occurred in one patient of Group II (10%), and in no patient of Group I (0%). the case of post-operative bleeding treated conservatively on the same day with control of bleeding by either catuerization or packing and it was successful.

The effects of PRP on bone graft are of beneficial issue. It has been reported that PRP improving bone regeneration in bone grafting, also maturity of grafted bone combined with PRP is significantly greater than that without PRP, and that grafted bone combined with PRP shows a mature Harversian system and a greater proportion of lamellar phase **[20]**. We did alveolar grafting during mixed dentition like Boyne who combined his work with orthodontic treatment in order to contribute to maxillary stability, allow dental eruption thought the grafted bone and facilitate posterior orthodontic treatment **[19]**.

The present study shows that CBCT scoring method for assessing alveolar bone graft outcomes indicated very good pre-and post-operative evaluation. Additionally, CBCT provides good accuracy for quantitative analyses of buccal and lingual alveolar bone thickness at different vertical levels.

Feichtinger had recommended using three-dimensional CT (3D CT) to obtain specific results on the volume and width of the bone bridge. Radiation exposure of pediatric patients with CT scan is much higher as compared to intraoral radiographs and the costs of 3D CT are greater than for CBCT **[20]**. In this study, we used CBCT, which have several benefits as: Radiation exposure to patients is less than for 3D CT examination methods; low cost, accurate assessments, longitudinal and transverse changes in grafted bone are correctly compared.

Most previous studies used CBCT to evaluate bone graft by comparing post-operative grafted bone volume with pre-operative volume of alveolar bony defect measured the volume of the pre-operative bony defect and post-operative bone fill **[21]**. Other previous studies similarly evaluated changes in the grafted bone after secondary alveolar bone graft by measuring the height, Labio-Lingual Thickness (LLT), and volume of the grafted bone at the time before bone graft compared with three and 12 months after bone graft. **[22]** many others also evaluated the resorption of alveolar bone grafting by assessing the graft volume at one month and six months **[23]**. The merit of these studies may be obscured because they all used one aspect of CBCT, in this study we use the most objective of CBCT.

In our study we found that PRP enhances bone density if used with iliac bone graft. As increase in bone mineral density, in PRP supported grafts has been reported ranging from 1.6 to 2.2 times that of a non PRP supported graft, as seen in this study. However, other studies, suggested that, it seems to be insufficient as a counter measure against bone resorption following secondary bone graft in the long term **[24]**. On conterary to another auther used supplemental demineralized bone matrix and allograft, and observed complete canine eruption in most of cases when combined with PRP **[25]**.

Platelet rich plasma may increase density of osteoblast in rabbit maxillary bone graft, collagen and osteoblast in early stage of hard callus forming were both affected by PRP and eventually trigger osteogenesis in rabbit maxillary bone graft **[26]**.

Conclusion:

PRP enhances bone formation in alveolar clefts when admixed with autologous bone graft harvested from the iliac crest as it leads to early bone formation, increased bone density, decreases bone resorption, low infection rate and least post-operative complications.

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