HISTOLOGICAL EVALUATION OF THE IMPACT OF CONCENTRATED GROWTH FACTOR ON HEALING FOLLOWING THE SINUS FLOOR AUGMENTATION

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Abstract

Background: The role of CGF has been established in the formation of the bone around dental implants. It has been said to form bone either in combination with xenograft/allograft or alone by itself. It is also seen that CGF helps in the improvement of bone quality around dental implants.

Aim: The present clinical study was aimed to histologically evaluate the impact of CGF, a new generation platelet derivative, on healing following sinus floor augmentation during maxillary sinus lift surgical procedures.

Methods: The present study assessed 18 subjects with bilaterally complete or partially edentulous maxilla where implants were placed following the split-mouth protocol. After sinus membrane lift, CGF was placed on one side and bovine xenograft on the other side chosen randomly, followed by hematoxylin-eosin and alizarin red staining.

Results: After hematoxylin-eosin and alizarin red staining, it was seen that significantly higher bone formation was seen at the side where CGF was placed with 112.43±26.36% and 96.14±24.47%, respectively, compared to the side where bovine xenograft was placed with 64.97±24.98% and 60.14±16.37% respectively with $p<0.05$. Also, residual graft material was significantly lower with CGF compared to bovine xenograft with $p<0.05$.

Conclusion: The present study concludes that during the placement of dental implants and sinus lift surgical procedures, the use of concentrated growth factors is a reliable modality with higher bone formation and lesser residual graft material compared to commercial xenografts.

Keywords: CGF, dental implants, lateral window sinus lift, sinus floor augmentation

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INTRODUCTION

Recent advances in graft materials, dental implants, and related surgical procedures have improved the prognosis in the placement of dental implants, especially in the maxillary posterior region. Tatum, in the late 1970s, was the first one to introduce the lateral window sinus lift technique, which is continuously being modified since its publication in 1980 by Boyne.[1] To perform the lateral window sinus lift technique, CBCT (cone beam computed tomography) is first used to assess the anatomy of the sinus, followed by cavity preparation in the lateral sinus wall that provide enough bone to place the dental implant of the standard length. To place an adequate dental implant, the Schneiderian membrane is detached from the wall of the sinus bone with appropriate instruments and is lifted under the membrane, and the space created is maintained with the graft materials.[2]

To lift the maxillary sinus floor, different graft materials are being used, namely alloplasts, allografts (both demineralized and mineralized), xenografts, and/or autologous bone. Also, platelet derivatives, including the PRF (platelet-rich fibrin), have been widely used in sinus lift surgeries as it is a high source of various growth factors resulting in better healing. There are lesser chances of implant survival when PRF is used alone for the sinus lift, and implant placement is simultaneous. However, good results have been reported when PRF is used combined with allografts during sinus lift procedures.[3]

In comparison to PRF, CGF (concentrated growth factors), another platelet derivative, has denser and larger fibrin matrices with more robust network structures, which make them highly effective in posing better osteogenesis. Also, CGF has a quick supply and is economical, making it a better alternative to PRF during the sinus augmentation procedure. Also, CGF decreases the risk of infection and increases the probability of regeneration as it is rich in stem cells CD 34+, white blood cells, and platelets.[4] CGF also releases various growth factors such as TGF (transforming growth factor), PDGF (platelet-derived growth factor), VEGF (vascular endothelial growth factor), and IGF (insulin-like growth factor). CGF has also shown the ability of bone regeneration in subjects with osteoporosis.

It has also been shown that sinus floor lifts using osteotomy with CGF followed by placement of short dental implants in subjects with severe atrophy of the maxilla have shown acceptable results, whereas the height of alveolar bone was found to be decreased after six months. However, bone loss in the next six months was insignificant statistically.[5] The role of CGF has been established in promoting bone formation around dental implants, either combined with xenografts or allografts or alone by itself, along with improved bone quality around dental implants.[6] The present clinical study was aimed to histologically evaluate the impact of CGF, a new generation platelet derivative, on healing following sinus floor augmentation during a maxillary sinus lift surgical procedure. The study also assessed the effect of CGF on the percentage of newly formed bone compared to pre-existing bone and the amount of fibrous connective tissue compared to the xenograft group.

MATERIALS AND METHODS

The present split-mouth clinical study was aimed to histologically evaluate the impact of CGF, a new generation platelet derivative, on healing following sinus floor augmentation during a maxillary sinus lift surgical procedure. The study also assessed the effect of CGF on the percentage of newly formed bone compared to pre-existing bone and the amount of fibrous connective tissue compared to the xenograft group. An informed consent in both written and verbal format was taken from all the subjects before study participation.

The study included 18 subjects from both genders and the age range of 35-75 years having posterior edentulous maxilla bilaterally (either partial or complete). In all 18 subjects included, one side was kept in the control group after random selection, where bovine xenograft was placed, and the other side was taken as a test group where CGF was placed. The inclusion criteria for the study were subjects having bilateral edentulous posterior maxilla with a remaining alveolar bone height of less than five mm between the sinus floor and alveolar crest. The exclusion criteria for the study were pregnancy, smoking, untreated, periodontal disease, untreated periapical disease (in sinus), use of bisphosphonates, immunosuppressant use, steroid use, autoimmune therapy, radiotherapy in head and neck region, malignancy, cardiovascular diseases, uncontrolled diabetes, malignant/benign sinus tumor, chronic/acute inflammatory diseases, cystic lesion, and any other signs and symptoms of sinus pathology.

CGF was prepared immediately before surgery with a collection of blood in 4-6 test tubes of 9 mm without anticoagulants (intravenous blood) but a clot activator, silicate, which was centrifuged following Mijiritsky E et al. [7] in 2021 as 30 seconds for acceleration followed by 2800 rpm, 2400 rpm, 2700 rpm, and 3000 rpm for 2, 4, 4, and 3 minutes respectively, and lastly 30-second speed reduction was made before the full stop. The complete rotation period was 14 minutes. Following centrifugation, the blood could be seen divided into four linings as red blood cells (RBCs) at the bottom, stem cell and growth factor layer of CGF, buffy coat/second layer, and serum layer at the top. The CGF clot was removed and separated from the RBC. All the surgical procedures were done by a single surgeon expert in the field. The surgery was done under local anesthesia following aseptic preoperative protocols.

For the surgery, following the crestal incision, a full-thickness mucoperiosteal flap was raised, and a lateral window sinus lift technique was adopted. After outlining the window, carbide round bur No.8 was used. This was followed by erosion to remove the window bone, and the Schneiderian membrane was elevated gently to prevent perforation from xenograft in controls and prevent collapse in the CGF group. For the control group, an acellular dermal matrix (ADM) was mixed with sterile saline and placed in the cavity. The subjects were advised for amoxicillin for one week at 8-hour intervals.

After Schneiderian membrane lifting, bovine xenograft was placed on one side randomly, and CGF was placed on the other side. After six months of placement, CBCT was done for both groups. This was followed by identifying the osteotomy area with a 2.7 mm diameter trephine bur. The samples were collected for histology, labeled, and placed in a container of 10% formalin which was sent for histologic examination.

This was followed by alizarin red and hematoxylin-eosin staining, followed by a microscopic assessment of the slide under 40 X magnification. The percentage of newly formed bone was reported as the ratio of new bone to former bone. The amounts of formed fibrous connective tissue, remaining graft material, and formed living bone were assessed and measured in mm2. The examining histologist was not aware of the sample and the two groups. The data gathered were analyzed statistically, and results were formed.

RESULTS

The present split-mouth clinical study was aimed to histologically evaluate the impact of CGF, a new generation platelet derivative, on healing following sinus floor augmentation during a maxillary sinus lift surgical procedure. The study also assessed the effect of CGF on the percentage of newly formed bone compared to pre-existing bone and the amount of fibrous connective tissue compared to the xenograft group.

It was seen that for alizarin red in test group staining before surgery, the mean bone was 95.81±43.74, which increased significantly to 197.02±72.57 at six months postoperatively with $p<0.01$. Similar results were seen for the control group, where bone formation improved significantly at six months to 169.73±87.32 compared to 99.41±37.32 before surgery with $p<0.01$. However, the difference in the test and control groups was statistically non-significant at baseline and six months, with p=0.832 and 0.483, respectively. The formed area was also significantly higher in the test and control groups, with 10.19±31.71 and 70.26±52.56, respectively, and $p<0.01$ for both. The difference in the formed area in the test and control group was non-significant with p=0.153, as shown in Table 1.

Staining	Number (n)	Before surgery (Mean ± S.D)	Six months after surgery (Mean ± S.D)	p-value	Formed bone area	p-value
Alizarin red						
Test	18	95.81±43.74	197.02±72.57	<0.01	10.19±31.71	<0.01
Control	18	99.41±37.32	169.73±87.32	<0.01	70.26±52.56	<0.01
p-value		0.831	0.483		0.153	
Hematoxylin-eosin						
Test	18	13.03±6.54	26.59±15.02	<0.01	13.59±8.68	<0.01
Control	18	13.92±8.94	21.84±13.13	<0.01	7.91±4.38	<0.01
p-value		0.834	0.463		0.109	

Table 1: Bone area (mm2) preoperative and six months postoperative in 2 groups of study subjects

Similar results were seen for the hematoxylin-eosin staining, where a significant increase in bone formation was seen in the test group with CGF from 13.03±6.54 pre-surgically to 26.59±15.02 at six months postoperatively with $p<0.01$. In the control group, a significant increase

was seen from 13.92±8.94 pre-surgically to 21.84±13.13 at six months postoperatively with p<0.01. The difference between the two groups at baseline and six months postoperative was statistically non-significant, with p=0.834 and 0.463, respectively. The area formed was significant in the control and test group, with p<0.01 for both (Table 1).

The study results showed no significant difference in the connective tissues formed in either test or control group with either hematoxylin-eosin or alizarin red staining. The connective tissue formed in the test group was 0.37±0.49 and 2.73±5.07 with hematoxylin-eosin and alizarin red stain, respectively, whereas, in the control group, the connective tissue was 0.52±0.56 and 2.27±4.73 with hematoxylin-eosin and alizarin red stain respectively. The p-value for hematoxylin-eosin and alizarin red was 0.153 and 0.466, respectively, as depicted in Table 2.

Parameter	**Hematoxylin eosin**	**Alizarin red**
Connective tissue		
Test	0.37±0.49	2.73±5.07
Control	0.52±0.56	2.27±4.73
p-value	0.153	0.466
Remaining material		
Test	0.000±0.00	0.000±0.00
Control	0.24±0.14	41.64±35.53
p-value	0.001	0.002
Formed bone		
Test	96.18±24.47	112.43±26.36
Control	60.18±16.37	64.97±24.98
p-value	0.002	0.001

Table 2: The percentage of connective tissue, remaining material, and formed bone with the two staining techniques used in the study

No remaining material was seen with hematoxylin-eosin stain in the test group, whereas it was 0.24±0.14 in the control group. The difference was statistically significant, with p=0.001. With Alizarin red stain, a significantly higher remaining graft was seen in the control group with 41.64±35.53, and no remaining graft material was seen in the test group. The difference was statistically significant with p=0.002 (Table 2).

On hematoxylin-eosin staining, formed bone was significantly higher in the test group at 96.18±24.47 compared to a control group where it was 60.18±16.37 with p=0.002. On alizarin red staining, the bone formed in the control group was 64.97±24.98, which was significantly lesser when compared to the test group, where it was 112.43±26.36 with p=0.001, as shown in Table 2.

DISCUSSION

The study results showed that for alizarin red in test group staining before surgery, the mean bone was 95.81±43.74, which increased significantly to 197.02±72.57 at six months postoperatively with p<0.01. Similar results were seen for the control group, where bone formation improved significantly at six months to 169.73±87.32 compared to 99.41±37.32 before surgery with p<0.01. However, the difference in the test and control groups was statistically non-significant at baseline and six months, with p=0.832 and 0.483, respectively. The formed area was also significantly higher in the test and control groups, with 10.19±31.71 and 70.26±52.56, respectively, and p<0.01 for both. The difference in the formed area in the test and control groups was non-significant, with p=0.153. These results were comparable to the studies of Chen X et al.[8] in 2018 and Kim TH et al.[9] in 2014, where histologic examination revealed significantly better bone formation from CGF in the atrophied maxilla.

It was also seen that for the hematoxylin-eosin staining where a significant increase in bone formation was seen in the test group with CGF from 13.03±6.54 pre-surgically to 26.59±15.02 at six months postoperatively with p<0.01. In the control group, a significant increase was seen from 13.92±8.94 pre-surgically to 21.84±13.13 at six months postoperatively with p<0.01. The difference between the two groups at baseline and six months postoperative was statistically non-significant, with p=0.834 and 0.463, respectively. The area formed was significant in the control and test group, with p<0.01 for both. These results were similar to the previous findings of Galindo-

Moreno P et al.[10] in 2018 and Sousa S et al.[11] in 2013, where authors reported a significantly higher increase in bone when CGF was used.

No significant difference was seen in the connective tissues formed in either test or control group with either hematoxylin-eosin or alizarin red staining. The connective tissue formed in the test group was 0.37±0.49 and 2.73±5.07 with hematoxylin-eosin and alizarin red stain, respectively, whereas, in the control group, the connective tissue was 0.52±0.56 and 2.27±4.73 with hematoxylin-eosin and alizarin red stain respectively. The p-value for hematoxylin-eosin and alizarin red was 0.153 and 0.466, respectively. These results were consistent with the previous studies of Kim BJ et al.[12] in 2021 and Sul SH et al.[13] in 2008, where authors suggested similar connective tissue formation in sinus lift surgical procedures.

The study results showed that no remaining material was seen with hematoxylin-eosin stain in the test group, whereas it was 0.24±0.14 in the control group. The difference was statistically significant, with p=0.001. With Alizarin red stain, a significantly higher remaining graft was seen in the control group with 41.64±35.53, and no remaining graft material was seen in the test group. The difference was statistically significant, with p=0.002. These results were in agreement with the findings of Kim HR et al.[14] in 2010 and Riben C et al.[15] in 2012, where authors reported no remaining material following bone augmentation in sinus lift surgical procedures.

Concerning the bone formed, on hematoxylin-eosin staining, formed bone was significantly higher in the test group with 96.18±24.47 compared to a control group where it was 60.18±16.37 with p=0.002. On alizarin red staining, the bone formed in the control group was 64.97±24.98, which was significantly lesser when compared to the test group, where it was 112.43±26.36 with p=0.001. These results were in line with the findings of Thor A et al.[16] in 2007 and Tekin U et al.[17] in 2019, where authors suggested higher bone formation with platelet concentrates and growth factors compared to bone graft only.

CONCLUSION

Considering its limitations, the present study concludes that during the placement of dental implants and sinus lift surgical procedures, the use of concentrated growth factors is a reliable modality with higher bone formation and lesser residual graft material compared to commercial xenografts. The study had limitations of a smaller sample size and shorter follow-up period.

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