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Randomized, controlled clinical trial to evaluate efficacy of sticky bone and concentrated growth factor in the management of intrabony defects: 12 months follow-up study

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ABSTRACT

Background: Platelet derivatives are enriched growth factors that ameliorate various cellular processes in regeneration. The present clinical trial aimed to evaluate and compare the effects of sticky bone and concentrated growth factors (CGFs) in the treatment of intrabony osseous defects by cone-beam computed tomography (CBCT).

Materials and Methods: The study included 20 patients having 40 intrabony defects. 20 sites each were included in both test group (Sticky bone) and Control group (CGF alone). The clinical parameters including probing pocket depth (PPD) and clinical attachment level (CAL) were assessed at baseline and 6 and 12 months posttherapy. The radiographic parameters including the depth, mesiodistal (MD), and the buccolingual (BL) width of the defect to assess the amount of bone fill were examined at baseline and after 12 months using CBCT.

Results: Twelve months posttherapy clinical results indicated a significant reduction of PPD and gain in CAL in both the study groups. Similar observations were recorded with CBCT radiographic parameters where the intrabony defect depth and MD defect width for the test group and control group significantly reduced after 12 months' posttherapy ($P < 0.0001$). However, no significant reduction in BL defect width was observed in control group ($P = 0.577$) in contrast to the test group ($P = 0.028$) after 12 months' posttherapy.

Conclusion: Intrabony defects treated with sticky bone showed improved clinical and radiographic parameters indicative of enhanced periodontal regeneration as compared to CGF alone treated sites.

Key Words: Concentrated growth factors, intrabony defects, periodontal regeneration, periodontitis, sticky bone

INTRODUCTION

Periodontitis is a complex inflammatory disease involving the combined effects of risk factors like environment, host, and bacteria.[1] One of the most complex processes of wound healing is potentially the regeneration of damaged periodontal tissue. Current clinical practice requires the surplus use of bio-materials, including bone derivatives and bone substitutes, guided tissue regeneration and biological factors such as enamel matrix proteins for the treatment of intrabony defects of one, two, and three walls or combination defects.[2,3]

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An innovative component of regenerative medicine is the concentrated growth factor (CGF). It is known to be the third-generation platelet concentrates prepared using continuous differential centrifugation. CGF contains more growth factors and has a harder fibrin structure and thicker elasticity than first-generation Platelet-rich plasma (PRP) and second-generation platelet-rich fibrin (PRF). The modification of the centrifugation rate enables the isolation of a larger and thicker fibrin matrix rich in growth factors, platelets, undifferentiated cells, leukocytes and CD34+, etc. This matrix of CGF aids in wound healing. It also contains immunological cells that help regulate inflammation and decrease the risk of infection. CGF is known to promote osteogenic differentiation and cell proliferation, thereby enabling bone formation; tissues healing and improving the quality of the newly formed bone.[4]

Sticky bone is a novel form of graft material that is also known as an enriched bone graft matrix with growth factors, prepared using autologous fibrin glue (AFG). AFG acts as a scaffold for migrating fibroblast as well as a hemostatic barrier. It induces and encourages angiogenesis and stimulates mesenchymal cells. It gives stability to the bone graft material within the defect and has the property of biocompatibility and biodegradability. This facilitates tissue healing and decreases bone resorption during the healing period.[5,6] The literature has shown little evidence describing the effectivity of sticky bone and CGF in the treatment of intrabony defects. Hence, the present study aimed to evaluate the effect of AFG enriched matrix (sticky bone) and CGFs in the treatment of intrabony osseous defects by cone-beam computed tomography (CBCT). The hypothesis of this study was that the AFG enriched bone graft matrix (sticky bone) was more effective than the CGFs in the treatment of intrabony osseous defects.

MATERIALS AND METHODS

Study design

The present study is a double-blind randomized controlled clinical trial conducted at the Department of Periodontics and Implantology of our institute. After getting approval from the institutional ethics committee the study was performed in accordance with the Helsinki declaration of 1975, as revised in 2013. The study was registered with the clinical trial registry-India (Clinical Trials Registry– India, CTRI/2018/10/016146). The study included patients between the age range 20–45 years with a mean age of 39 years. Forty intrabony defects were selected from 20 patients (9 males and 11 females) with Stage III (Grade A) periodontitis, classified according to consensus report of workgroup 2 of the 2017 World workshop classification of periodontal disease.[7] The randomization and plan of work are explained in Figure 1. All patients were informed about the procedure being performed and written consent was obtained from them.

Comprehensive case history, clinical examination, radiographic assessment, and periodontal indices of the patient were noted to have a systematic and organized recording of observations and information.

Patients with probing pocket depth (PPD) ≥6 mm and clinical attachment level (CAL) ≥5 mm with the minimum of two osseous defects (either two or three walled) with a depth of ≥3 mm whose architecture had to be confirmed by direct observation during surgical exposure were included. Patients showing radiographic evidence of intrabony defects as initially revealed by radiovisiographs and later on confirmed and standardized by CBCT were selected.

Patients with systemic diseases, allergies, or drug usage, with a history of periodontal treatment in the previous 6 months and pregnant/lactating women were excluded from the study.

Nonsurgical periodontal therapy

Nonsurgical periodontal therapy was given to the patients followed by oral hygiene instructions. The patients were re-evaluated 6 weeks after initial therapy to assess the status of complete oral hygiene. Then, the selected sites were randomly assigned as test group (sticky bone) and control group (CGF) by computer-generated software.

Clinical and radiographic examination

The surgical procedure was carried out by a single operator discontinuous Galerkin and the clinical parameters were noted by the assessor (SR) who was blinded to the procedure. The clinical parameters involving plaque index (PI),[8] gingival index (GI),[9] PPD, and CAL were recorded using periodontal probe‡ at baseline after 6 and 12 months. Custom-made occlusal acrylic stents with grooves were made to standardize the probe angulation and position.

Intrabony defects were assessed using CBCT§ at baseline and after 12 months follow-up. The depth

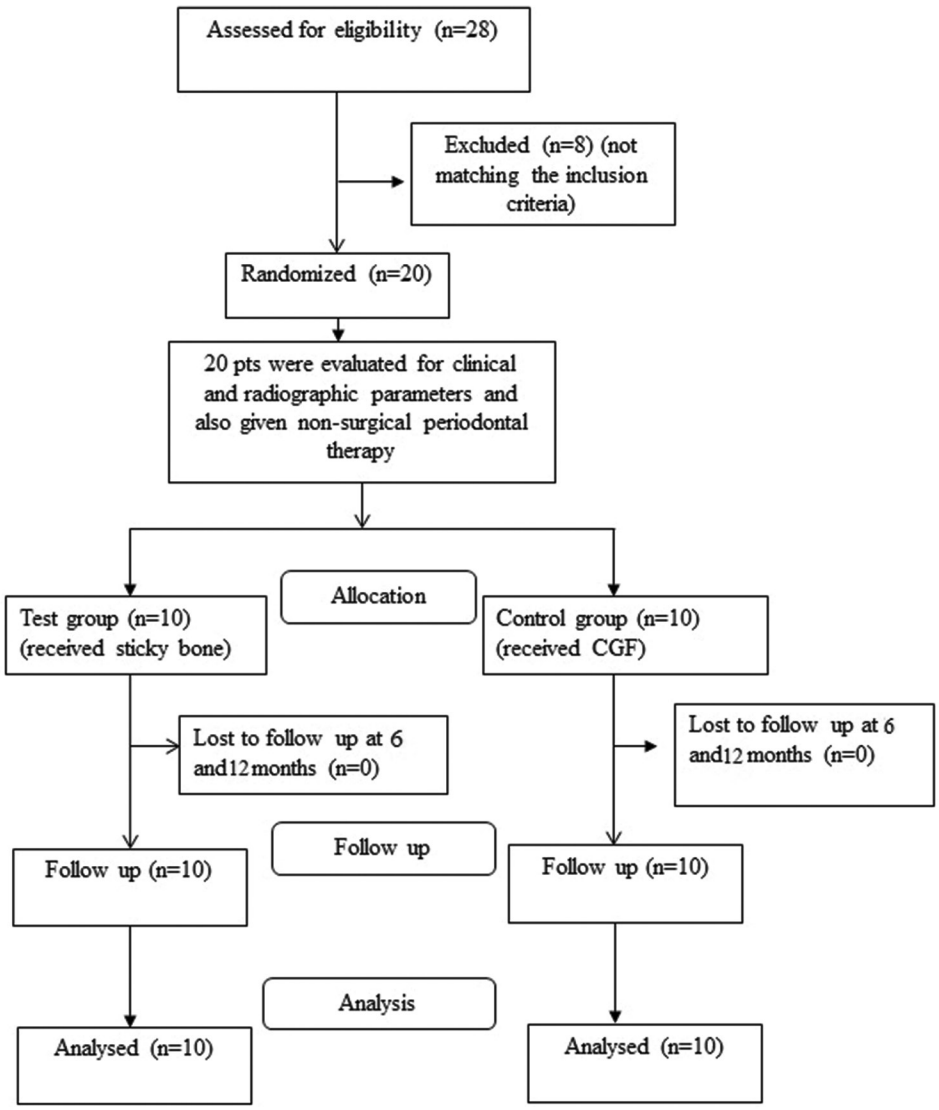

Figure 1: Plan of work.

of the defect was measured as distance from the alveolar crest to base of the defect (AC), whereas, the Mesiodistal (MD) distance was recorded from AC-AC' as shown in Figure 2a. Buccolingual (BL) bone defect width was measured from B-L analyzed by CBCT as shown in Figure 2b. The lowest discontinuous point of the periodontal ligament was considered as the landmark of the base of the defect.

Preparation of concentrated growth factor

CGF was prepared according to the protocol given by Rodella *et al*.[10] 9 ml of blood was drawn in a sterile test tube without anticoagulant. These tubes were then immediately centrifuged†. The type of Centrifuge used was Table Top single Phase Remi Compact Laboratory Centrifuge R 4C, using specific centrifugation protocol as follows: 30 s-acceleration, 2 min–2700 rpm, 4 min-2400 rpm, 4 min–2700 rpm, 3 min 3000 rpm, 36 s-deceleration, and stop. At the end of the process, three blood layers were obtained: The upper layer containing platelet-poor plasma, the middle layer with fibrin-rich gel with aggregated platelets, and CGFs and the lower layer comprised red blood cell (RBC).[10]

Preparation of sticky bone

Sticky bone was formulated according to the protocol given by Sohn *et al*.[5] 10 ml of venous blood was drawn from the antecubital fossa of the patient and collected in the test tubes. Noncoated test tubes having red-colored caps were used. It was then centrifuged at 2400–2700 rpm. The centrifugation time for AFG varies from 2 to 12 min. The centrifuge† was stopped after 2 min. AFG obtained was separated from the test

tube. The noncoated tube showed 2 different layers. The upper layer comprised of AFG while the bottom layer consisted of the RBC which was discarded. The upper layer of AFG was collected with the syringe and mixed with particulate bone powder and allowed for 5–10 min for polymerization to produce yellow-colored sticky bone.[5]

Surgical periodontal therapy

An intraoral asepsis was performed by preprocedural mouth wash by 10 ml of 0.12% chlorhexidine gluconate. The extraoral asepsis was carried out by swabbing with 5% povidone-iodine. After the administration of local anesthesia#, intra-sulcular incision extending at least one tooth mesial and distal to the surgical site preserving the interdental papilla wherever possible was given and a full-thickness mucoperiosteal flap was reflected. After the complete debridement of intrabony defects, the CGF was placed in the defect site in the control group while the sticky bone was placed in the test group [Figure 3]. Immediately after placing the CGF and sticky bone, the reflected flap was approximated and repositioned. The interrupted sutures ** and periodontal dressing ††were placed at surgical sites.

Postoperative care

Postoperative instructions and antibiotics ‡‡ and analgesic §§ were prescribed twice daily for 7 days. The patients were advised Chlorhexidine oral rinse (10 ml twice daily) and refrained from chewing hard and sticky foods. In addition to this, they were instructed and motivated to use toothbrush with soft bristles for the next 12 months.

Postsurgical measurements

The clinical parameters were evaluated at baseline, 6, and 12 months follow-up, whereas radiographic parameters were recorded at baseline and 12 months follow-up using CBCT. Figure 4 shows baseline and 12 months CBCTs for the Test and the Control group.

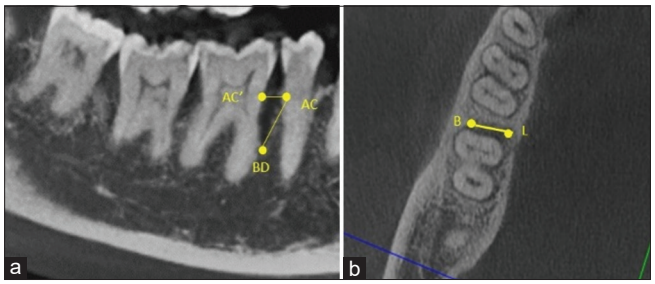

Figure 2: Schematic of reference points for measurement of Cone Beam Computed Tomography parameters. (a) Defect depth and Mesiodistal Width. (b) Buccolingual Width.

Data analysis

The sample size was calculated with reference to the result obtained from the study by Qiao *et al*.[6] The data on the clinical trial for intrabony defect regeneration was referred. Estimation power analysis was performed to obtain the sample size. The data for the quantity of defects from patients resulted in an effect size of 0.7. To accomplish the stringent effect with 90% power and 95% confidence level, a sample size of 40 intrabony defects was essential.

The clinical and radiographic recordings were also summarized in terms of mean and standard deviation for each time point and each site. The comparison of the mean difference between sites, at each time point, was carried out using paired *t*-test, while the comparison across times, for each site, was carried out using repeated measure SPSS ver 20.0 (IBM Corp., ARMONK USA) software was used for all the analyses and the statistical significance was tested at 5% level.

RESULTS

A total of 20 patients were treated with CGF + PRF and none of them reported of discomfort or adverse effect and uneventful healing during the study. Table 1 provides the comparison of clinical parameters, i.e., PI and GI across time in the study sample. Statistically significant changes in the mean PI were found. from baseline to 12 months, with $P < 0.001$ at the test site. As far as the GI is concerned, the mean changed from 1.57 ± 0.25 at baseline to 1.21 ± 0.15 after 6 months and 0.97 ± 0.31 after 12 months, with the $P < 0.0001$.

The comparison of PPD and CAL between two groups as well as comparison across time for each site is shown in Table 2. The mean differences

Table 1: Comparison of plaque index and gingival index across time

Parameters	*n*	Mean±SD	*P**
PI			
Baseline	20	3.40±0.38	<0.0001 (S)
6 months	20	2.48±0.49	
12 months	20	1.71±0.69	
GI			
Baseline	20	1.57±0.25	<0.0001 (S)
6 months	20	1.21±0.15	
12 months	20	0.97±0.31	

*Calculated using repeated measure ANOVA, *P*<0.0001 was considered as significant. *n*: Number of patients; S: Significant; SD: Standard deviation; PI: Plaque index; GI: Gingival index

in the PPD values at baseline between the two groups were statistically insignificant (P = 0.999). Similarly, the mean PPD for the Test group and the control group at 6 months was 4.69 ± 0.82 mm and 4.80 ± 1.11 mm respectively (P = 0.447) while after 12 months it was 3.20 ± 0.70 and 3.60 ± 0.99 mm respectively (P = 0.042). CAL values for both groups at baseline did not show significant differences (P = 0.359). After 6 months, mean CAL was found to be statistically insignificant (P = 0.301) when compared in both the groups. However, it was statistically significant after 12 months (P = 0.002). There was more CAL gain in the test group as compared to the Control group.

Table 3 depicts CBCT analysis at baseline and after 12 months of bone defect depth, MD and BL width. After 12 months, significant differences were found in both the groups with a defect depth of 2.69 ± 0.49 mm in the test group and 3.20 ± 0.88 mm in the control group. However, the differences were statistically insignificant in the MD width among both the groups with values of 3.12 ± 0.59 mm in the test group and 3.24 ± 0.82 mm in the cntrol group. The values of MD width after 12 months between the test and the control group were 2.41 ± 0.42 mm for the Test group and 2.80 ± 0.75 mm for the control group (P = 0.037). The BL width at baseline for the Test group was 2.95 ± 0.73 and for the control group was 3.35 ± 0.86 mm which was statistically insignificant. The reduction in the test group was 2.59 ± 0.54 mm while that in the control group was 3.18 ± 1.21 mm after 12 months which was statistically significant (P = 0.037).

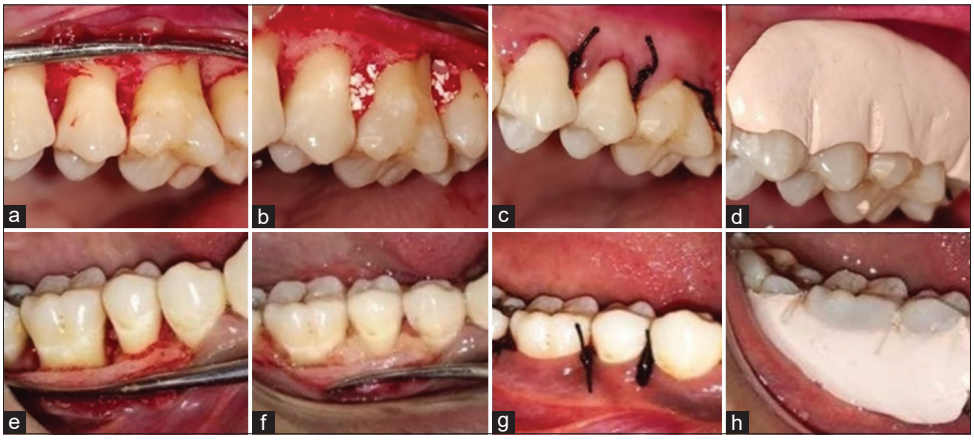

Figure 3: Surgical procedure for Test Group and Control Group. (a) Reflection of flap for Test Group. (b) Placement of Sticky Bone in defects for Test Group. (c) Suture placement for Test Group. (d) Periodontal Pack Placement for Test Group. (e) Reflection of flap for Control Group. (f) Placement of Concentrated Growth Factors in defects for Control Group. (g) Suture placement for Control Group. (h) Periodontal Pack Placement for Control Group.

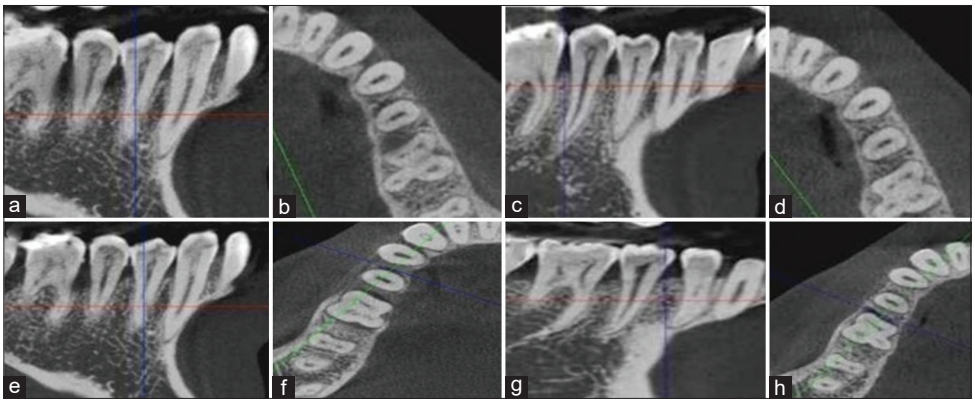

Figure 4: Cone Beam Computed Tomography for Test Group and Control Group. (a) Sagittal view at baseline for Test Group. (b) Transverse view at baseline for Test Group. (c) Sagittal view at 12 months for Test Group. (d) Transverse view at 12 months for Test Group. (e) Sagittal view at baseline for Control Group. (f) Transverse view at baseline for Control Group. (g) Sagittal view at 12 months for Control Group. (h) Transverse view at 12 months for Control Group.

Table 2: Comparison of probing pocket depth and clinical attachment level in mm between two groups at each time point and across times in each group

Parameters	Mean±SD		*P**
	Test group (*n*=20)	Control (*n*=20)	
PPD (mm)			
Baseline	6.70±1.03	6.70±1.22	0.999 (NS)
6 months	4.60±0.82	4.80±1.11	0.447 (NS)
12 months	3.20±0.70	3.60±0.99	0.042 (S)
*P*ǂ	<0.0001 (S)	<0.0001 (S)	
CAL (mm)			
Baseline	7.30±1.17	7.50±1.24	0.359 (NS)
6 months	5.55±1.36	5.25±1.29	0.301 (NS)
12 months	3.20±0.89	4.15±1.04	0.002 (S)
*P*ǂ	<0.0001 (S)	<0.0001 (S)	

ǂCalculated using Repeated measure ANOVA; *Calculated using *t*-test for independent samples, *P*<0.0001 was considered as significant. *n*: number of patients; S: Significant; NS: Not significant; PPD: Probing pocket depth; CAL: Clinical attachment level; SD: Standard deviation

Table 3: Comparison of defect depth, mesiodistal, and buccolingual width in mm between two groups at each time point and across times in each group

	Mean±SD		*P**
	Test group (*n*=20)	Control group (*n*=20)	
Defect depth			
Baseline	3.92±0.84	3.95±0.95	0.919 (NS)
12 months	2.69±0.49	3.20±0.88	0.041 (S)
*P*ǂ	<0.0001 (S)	<0.0001 (S)	
Defect width (MD)			
Baseline	3.12±0.59	3.24±0.82	0.572 (NS)
12 months	2.41±0.42	2.80±0.75	0.037 (S)
*P*ǂ	<0.0001 (S)	<0.0001 (S)	
Defect width (BL)			
Baseline	2.95±0.73	3.35±0.86	0.101 (NS)
12 months	2.59±0.54	3.18±1.21	0.037 (S)
*P*ǂ	0.028 (S)	0.577 (NS)	

ǂCalculated using paired *t*-test; *Calculated using *t*-test for independent sample, *P*<0.0001 was considered as significant. *n*: number of patients, S: Significant; NS: Not significant; SD: Standard deviation; MD: Mesiodistal; BL: Buccolingual

DISCUSSION

The present study was a double-blinded randomized controlled trial assessing the efficacy of CGF and sticky bone for the treatment of periodontal intrabony defects. Platelet concentrates provide rich source of growth factors and therefore inhibit hemorrhage, tissue adherence, promote healing, minimize pain and accelerate the formation of new tissues.[11] AFG is a biological product prepared using patient's own blood and has benefits like decreased bleeding, reduced scarring, and serous fluid collection.[5]

A very limited literature is available on the use of sticky bone in the regeneration of intrabony defects. In this study, therefore the effectiveness of sticky bone in the treatment of periodontal intrabony osseous defect was evaluated. Each group consisted of the equal number of 2 walled and 3 walled defects. The histological assessment is preferred approach for the evaluation of regeneration but because of the invasive nature of evaluation and ethical issues, CBCT was used in the current study with a more detailed radiographic technique.

Sticky bone has tremendous benefits in periodontal regeneration. It has mouldable nature and a strong interlinked fibrin network due to which it can be well adapted in different shapes of bony defects. As a result of entrapment of platelets and leukocytes in the fibrin network to release the growth factor, it accelerates regeneration of bone and this reduces the need of bone tack or titanium mesh. It minimizes soft tissue ingrowth due to sturdy fibrin interaction and also, no biochemical additives are required for its preparation.[5] The mineral scaffold contains bone cells that are needed for the formation of bones and growth factors that are necessary for cell stimulation.[12,13]

On the other hand, the main role of CGF is that it increases osteoblast development and bone repair, which speeds up osseointegration. Angiogenesis and tissue remodeling are aided by CGF, which contains fibrinogen, growth factors, leukocytes, coagulation factors, endothelial growth factors, and platelets. CGF also reduces scarring due to its numerous advantages like homeostatic and tissue healing properties, promotes wound healing and osteogenesis, accelerates epithelial, endothelial, and epidermal regeneration. Due to the high concentration of leukocytes, it has high antimicrobial properties and provides scaffold for supporting cytokines and cellular migration.[4]

In the present study, the PPD reduction was statistically significant in both the groups at 6 and 12 months compared to baseline. Nevertheless, PPD reduction was more in the Test group as compared to the control group at 12 months follow up. These results were in accordance with the study performed by Juneja and Bharti.[11] who evaluated and compared the combination of PRF combined with alloplastic bone graft that is Demineralised Freeze-Dried bone graft, Hyaluronic acid (HA), and HA alone outcomes in the treatment of intrabony defects.

In the present study, statistically significant difference was found with respect to MD and BL width reduction

in the test group as compared to the control group which was in agreement with the study by Bodhare *et al*.[14] were they reported the significant increase in CAL gain and reduction in BL and MD dimensions of bone defect in bioactive glass + PRF Group as compared to without PRF Group. Furthermore, similar results were found in studies done by Pajnigara *et al*.[15] and Shah and Kolte.[16]

The present study used CBCT to look for the reduction in the defect depth as used by Bodhare *et al*.[14] A statistically significant reduction in defect depth between the two study groups was found. These results are in accordance with the study done by Wanikar *et al*.[17] in the treatment of grade II furcation defect on use of 1% alendronate (ALN) gel combined with PRF and PRF alone and a statistically significant reduction in PPD and CAL gain was more in 1% ALN gel combined with PRF. PRP + β-tricalcium phosphate (β TCP) and β TCP alone were used in another study done by Saini *et al*.[18] where they found more defect depth reduction with PRP + β TCP as compared to β TCP alone.

Promoting tissue healing, reduction of the bone loss throughout the healing period and steadiness of sticky bone in the defect site are the possible reasons for getting the improved results in the test group as compared to the Control group. However, the present study has some limitations as the sample size is small, better results would have been achieved with a larger sample size. Moreover, the gender-based evaluation would be required to substantiate the results and to govern the stability of the outcomes long-term analysis is required.

CONCLUSION

Within the confines of the study, it can be concluded that the sticky bone is more effective in gaining the CAL, reduction in PPD and radiographic outcomes like defect depth as compared to CGF alone. Thus, can be preferred over CGF for the treatment of intrabony osseous defects. Furthermore, the use of CBCT proved to be a better replacement over the invasive histologic evaluation.

Footnotes for manuscript

- ‡ UNC-15 Periodontal Probe, Hu-Freidy, Chicago, IL, United States
- § The Orthophos® XG 3D manufactured by Sirona Dental Systems GmbH, Germany
- † R-4C, REMI, Mumbai, India
- Dentrox, MonarkBiocare Private Limited Chandigarh, India
- # Lox 2% adrenaline (1:200000), Neon Laboratory Limited, Mumbai, India
- ǂ Gracey curettes, Hu-Freidy, Chicago, IL, United States
- *3-0 Ethicon Mersilk, Somerville, New Jersey, United States
- †† GC coepak periodontal dressing, Alsip, IL, USA
- ‡‡Moxikind CV 625 mg, Mankind Pharma, New Delhi, India
- §§Ketorolac tromethamine10 mg, Dr. Reddys Laboratories, Hyderabad, India.

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Conflicts of interest

The authors of this manuscript declare that they have no conflicts of interest, real or perceived, financial or nonfinancial in this article.

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