BABU BANARASI DAS UNIVERSITY

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES, LUCKNOW

COMPARATIVE EVALUATION OF THE EFFECTIVENESS OF DEMINERALIZED FREEZE- DRIED BONE ALLOGRAFT(DFDBA) ALONE AND DEMINERALIZED FREEZE-DRIED BONE ALLOGRAFT(DFDBA) WITH RECOMBINANT HUMAN BONE MORPHOGENETIC PROTEIN-2(rhBMP-2) IN THE TREATMENT OF INTRABONY DEFECTS : A CLINICO - RADIOGRAPHIC STUDY

DISSERTATION

Submitted to

BABU BANARASI DAS UNIVERSITY,

LUCKNOW, UTTAR PRADESH

In the partial fulfilment of the requirements for the degree

of

MASTER OF DENTAL SURGERY

in

PERIODONTOLOGY

By

Dr. ANKIT BHADANI

Under the guidance of

Dr. SURAJ PANDEY

READER

Department of Periodontology

Batch 2020-2023

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DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled **COMPARATIVE EVALUATION OF THE EFFECTIVENESS OF DEMINERALIZED FREEZE- DRIED BONE ALLOGRAFT(DFDBA) ALONE AND DEMINERALIZED FREEZE-DRIED BONE ALLOGRAFT(DFDBA) WITH RECOMBINANT HUMAN BONE MORPHOGENETIC PROTEIN-2(rhBMP-2) IN THE TREATMENT OF INTRABONY DEFECTS : A CLINICO - RADIOGRAPHIC STUDY"** is a bonafide and genuine research work carried out by me under the guidance **Dr. SURAJ PANDEY** Reader, Babu Banarasi Das College of Dental Sciences, Babu Banarasi Das University, Lucknow, Uttar Pradesh.

Date: 4/02/23

Place: Lucknow

Dr. ANKIT BHADANI

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Dedicated to my Grandparents

Late Smt. Gayatri Devi Bhadani &

Late Shri Radha Krishna Bhadani

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DFDBA	Demineralized freeze- dried bone allograft
rhBMP-2	Recombinant human bone morphogenetic protein-2
FDBA	Freeze- dried bone allograft
BMP	Bone morphogenetic protein
TGF	Transforming growth factor-
PRP	Platelet rich plasma
RBC	Red blood cell
PRF	Platelet-rich fibrin
AATB	American Association of Tissue Banks
GI	Gingival index
PI	Plaque index
CAL	Clinical attachment level
PPD	Probing pocket depth
GTR	Guided tissue regeneration

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Place: Lucknow

Dr. Ankit Bhadani

ABSTRACT

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Aim: To evaluate and compare the clinical and radiographic outcomes observed in treating intrabony defects with DFDBA alone and DFDBA in conjunction with rhBMP2.

Materials and Method: A total of 20 patients fulfilling the inclusion and exclusion were randomly divided into 2 groups i.e. Group A (DFDBA alone) and group B (DFDBA + rhBMP-2). All the clinical parameters (PI, GI, PPD and CAL) and radiographic parameters were recorded at the baseline and after 6 months.

Result: PI, GI, PPD and CAL show no statistically significant difference in between the two groups. Bone gain shows statistically significant difference in between the two groups.

Conclusion: The 2 treatment modalities (Group A & B) showed favorable clinical results. DFDBA with rhBMP-2 showed better results in comparison with DFDBA alone. In our study we observed that the BMPs in DFDBA are somewhat in an inactive form and the addition of rhBMP-2 to DFDBA attained better results in terms of bone gain.

INTRODUCTION

INTRODUCTION

A persistent inflammatory condition called periodontitis damages the periodontal tissues and eventually leads to tooth loss. Cementum, periodontal ligament, alveolar bone, and gingiva are only a few of the tissues that must regenerate for periodontal repair to be successful. If left untreated, periodontitis, an infectious condition that damages the tooth-attachment system, results in a progressive loss of attachment and may finally cause early tooth loss.[1,2]. Periodontal therapy aims to return diseased periodontal tissues to their pre-disease architectural shape and function. This calls for the development of cementum, the regeneration of missing bone, the regeneration of the gingival connective tissues destroyed by inflammation, and the re-implantation of connective tissue fibers into previously infected root surfaces[3]. The periodontium ability to regenerate itself revived the desire to investigate other methods and materials for the purpose. Periodontists have a long-standing interest in replacing the tissues that support teeth that have been lost to periodontal disease[4].

Scaling, root planing, gingival curettage, gingivectomy, and other flap techniques, including osseous surgery, are all common forms of traditional periodontal therapy that are useful for stabilizing periodontal status and preserving periodontal health. The restoration of the periodontium to normal has frequently been a challenging goal of periodontal therapy. The primary outcome of this therapeutic approach has been periodontium repair as opposed to regeneration. Because it involves both calcified (the bone and cementum) and soft connective tissues (the gingiva and periodontal ligament), periodontal regeneration is unusual. In order for regeneration to take place, all periodontal components must be coordinated and integrated during their repair. Due in part to the intricacy of the biological events, variables, and cells underlying successful periodontal regeneration, complete regeneration may be an unattainable goal in many instances.[5,6].

To accomplish the worthwhile goal, a number of important difficulties that collectively operate as a roadblock in the way of total regeneration must be addressed. The purpose of this review is to inform the reader on the crucial topics relating to periodontal regeneration.

The goal of periodontal therapy is to reduce periodontal pockets for simple plaque control and to encourage the growth of new periodontal tissue where it is needed.[7]

The gold standard is an autogenous bone graft, although the practise of using it is limited due to additional surgery and side effects[8–11] Despite having osteoinductive properties, the allogenous bone grafts safety could not be established.[12] Both the synthetic bone graft and the xerogenous bone graft have osteoconductive properties but not osteoinductivity. According to Piorellini et al., these grafts couldn't be used successfully in graft surgery.

Demineralized freeze-dried bone allograft (DFDBA) implantation has a long history in periodontics. Periodontists have attempted to use the osteoinductive elements probably present in the graft for the promotion of periodontal bone regeneration since Urist's initial publications.[13,14] Indeed, commercially accessible bone preparations from several bone banks contained BMP-2, -4, and -7. The biological activity did seem to be lower than it was with fresh preparations[16], though, and there was a lot of variation in the osteoinductive qualities of various preparations, as opposed to fresh preparations,[15]. Additionally, Becker et al.[17] questioned the need for commercially accessible demineralized bone in periodontics after studying the osteoinductive capabilities of DFDBA. Instead, they asked that recombinant BMPs of known quality and quantity be loaded into a carrier matrix.

Mesenchymal stem cells and growth factors have recently been tested in the regeneration of periodontal tissue. In 1965[18], Urist reported on the impact of recombinant human bone morphogenetic protein (rhBMPs) of growth factors on bone regeneration.

The rhBMPs' DNA sequence and recombinant production method have been identified and established.[19–21] According to the converted osteoblast from myoblasts that was detected in the bone matrix saturated with rhBMP-2,[19,28] mice's muscles produced new isotope bone.

Because of their influence on osteoblast, chondroblast, and osteoclast, rhBMPs, a member of the TGF-beta (transforming growth factor-beta) superfamily, are crucial for the formation of the mammalian skeleton. [20,21]

Clinical trials[22–26] had established the role of rhBMP-2 in bone repair. According to animal studies, the effect of rhBMP-2 on the regeneration of periodontal tissue was favourable.[27–31]

No research has been done on how rhBMP-2 affects the regeneration of periodontal tissue in humans. In this clinical investigation, periodontal tissue regeneration in 2- and 3-wall intrabony periodontal defects will be compared to the effects of rhBMP-2 and allograft.

Due to the fact that DFDBA creates a strong bone foundation for the development of new bone, its use as an osteoconductive agent has had superior clinical success. However, little is known about how biologic osteoinductive agents like BMPs contribute to the osteoconductive process.

AIM AND OBJECTIVES

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AIM: To evaluate and compare the clinical and radiographic outcomes observed in treating intrabony defects with DFDBA alone and DFDBA in conjunction with rhBMP2.

OBJECTIVE:

- To evaluate the efficacy of DFDBA for treating vertical defects.
- To assess the efficacy of DFDBA combined with rhBMP-2 for treating vertical defects.
- To compare the difference in efficacy between the two groups.
- To assess the benefit of adding rh BMP-2 to DFDBA in treating intrabony defects

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REVIEW OF LITERATURE

REVIEW OF LITERATURE

1. **Mellonig J.T., Bowers G.M. and Cotton W.R. 1981**[32]conducted a study to make a direct histological comparison of new bone formation evoked by decalcified freeze-dried bone allograft, freeze-dried bone allograft, autogenous osseous coagulum, and autogenous bone blend. Defects were surgically created in the calvaría of 35 guinea pigs. The graft materials were placed in porous nylon chambers and implanted into the defects. Implanted empty nylon chambers served as controls. The animals were sacrificed at 3,7,14, 21, 28,35, and 42 days. New bone formation was determined quantitatively from histology preparations. It was concluded that, in this model system, decalcified freeze dried bone allograft is a graft material of high osteogenic potential; autogenous osseous coagulum and bone blend of less potential, and freeze-dried bone allograft even less.

2. **Toriumi D.M., Kotler H.S., Luxenberg D.P., Holtrop M.E. and Wang E.A. in 1991**[33] conducted a study on Bone morphogenetic protein\p=n-\2(BMP-2) is a human recombinant bone-inducing factor that stimulates bone formation within 14 days In group 1, reconstruction plates were removed at 10 weeks because stiff, non compressible mineralized bone formed across the defects, allowing the animals to chew a solid diet. The defects from groups 2 and 3 showed minimal, if any, bone formation and remained grossly unstable, prohibiting plate removal or advancement to a solid diet. The biomechanical strength of the defects reconstructed with BMP-2 increased significantly from 3 to 6 months and was related to degree of mineralization and thickness of bone bridging the defect.

3. **Anderegg C.R., Martin S.J., Gray J.L., Mellonig J.T., Gher M.E. 1991**[34] conducted a study to evaluate the potential of decalcified freeze-dried bone allograft (DFDBA) combined with a barrier material in the treatment of human molar furcation defects (experimental) as compared to the barrier technique alone (control). Fifteen pairs of Class II or III furcation invasion defects comprised the study group. Six months post-treatment, each site was surgically reentered and measurements repeated. Following either treatment, recession was minimal with statistically significant improvement in probing depth reduction and clinical attachment level gain favoring the combined technique. Hard tissue changes were comparable for alveolar crest

resorption, however, there was a distinct difference, statistically, for both horizontal and vertical bone repair favoring the use of the demineralized bone graft in combination with the e-PTFE membrane.

4. **Garraway R., Young W.G., Daley T., Harbrow D., Bartold P.M. in 1998**[35] studies conducted a study to demonstrated that implantation of laboratory preparations of demineralized freeze dried bone (DFDB) into the thigh muscle of mice induces ectopic osteoinduction. Histological analysis of the DFDB/collagen sponges demonstrated significant remineralization which increased with time. The results found for the DFDB/collagen sponge indicate a different mechanism of activity from DFDB as evidenced by its rapid remineralization.

5. **Schwartz Z., Somers A., Mellonig J.T., Carnes Jr D.L., Wozney J.M., Dean D.D., Cochran D.L., Boyan B.D. in 1998**[36] conducted a study on commercial preparations of human demineralized freeze-dried bone allograft (DFDBA) vary in their ability to induce new bone formation. This study tested the hypothesis that inactive DFDBA can be used as an effective carrier of recombinant human bone morphogenetic protein2 (rhBMP-2). Two batches of active DFDBA were used as controls. The results showed that active DFDBA induces new bone formation, whereas inactive DFDBA does not. Addition of rhBMP-2 to inactive DFDBA results in new bone formation with a bone induction index comparable to that of active DFDBA. Their study shows that addition of rhBMP-2 to inactive DFDBA provides reproducible, consistent bone induction, and suggests that inactive commercial preparations may contain inadequate amounts of BMP to cause bone induction compared to active preparations.

6. **Li H, Pujic Z, Xiao Y, Artold PM in 2000**[37] proposed a study that demineralized freeze-dried bone allograft (DFDBAs) as a useful adjunct in periodontal therapy to induce periodontal regeneration through the induction of new bone formation. The presence of bone morphogenetic proteins (BMPs) within the demineralized matrix has been proposed as a possible mechanism through which DFDBA may exert its biologic effect. However, in recent years, the predictability of results using DFDBA has been variable and has led to its use being questioned. One reason for the variability in tissue response may be attributed to differences in the processing of DFDBA, which

may lead to loss of activity of any bioactive substances within the DFDBA matrix. Therefore, the purpose of this investigation was to determine whether there are detectable levels of bone morphogenetic proteins in commercial DFDBA preparations. These results indicate that all of the DFDBA samples tested had no detectable amounts of BMP-2 and -4. In addition, an unknown substance present in the DFDBA may be responsible for degradation of whatever BMPs might be present.

7. **Jepsen S, Terheyden H in 2002**[38] performed a large numbers of studies over the last ten years, have demonstrated the possibility of periodontal tissue regeneration by bone morphogenetic proteins. There is evidence for the promotion of periodontal wound healing by rhBMP-2 and rhBMP-7 from multiple in vitro and preclinical trials. Provided human clinical trials confirm these findings and growth factor therapies receive approval by the health authorities, the therapeutic use of these potent biologics will certainly add to our regenerative clinical strategies.

8. **Jovanovic SA et al in 2003**[39] conducted a study to evaluate bone formation and BIC at long-term, functionally loaded, endosseous dental implants placed into bone induced by recombinant human bone morphogenetic protein-2 (rhBMP-2) in an absorbable collagen sponge (ACS) carrier. There were no significant differences between dental implants placed into rhBMP-2/ACS induced bone and resident bone for any parameter at any observation interval. They concluded that, rhBMP-2/ACS-induced bone allows installation, osseointegration, and long-term functional loading of machined, threaded, titanium dental implants in dogs..

9. **Jung RE, Glauser R, Schärer P, Hämmerle CH, Sailer HF, Weber FE in 2003**[40] conducted a study to test whether or not the addition of recombinant human bone morphogenetic protein-2 (rhBMP-2) to a xenogenic bone substitute mineral (Bio-Osss) will improve guided bone regeneration therapy regarding bone volume, density and maturation. In 11 partially edentulous patients, 34 Branemark implants were placed at two different sites in the same jaw (five maxillae, six mandibles) requiring lateral ridge augmentation. The bone defects were randomly assigned to test and control treatments: the test and the control defects were both augmented with the xenogenic bone substitute and a resorbable collagen membrane (Bio-Gides). It is concluded that the combination of the xenogenic bone substitute mineral with

rhBMP-2 can enhance the maturation process of bone regeneration and can increase the graft to bone contact in humans. rhBMP-2 has the potential to predictably improve and accelerate guided bone regeneration therapy.

10. **Xiao YT, Xiang LX, Shao JZ. in 2007**[41] conducted a study on bone morphogenetic proteins (BMPs) are multi-functional growth factors belonging to the transforming growth factor-beta super family. It has been demonstrated that BMPs had been involved in the regulation of cell proliferation, survival, differentiation and apoptosis. However, their hallmark ability is that play a pivotal role in inducing bone, cartilage, ligament, and tendon formation at both heterotopic and orthotopic sites. In this review, they mainly concentrate on BMP structure, function, molecular signaling and potential medical application.

11. **Lan J, Wang ZF, Shi B, Xia HB, Cheng XR in 2007**[42] investigated a study whether osseointegration can be enhanced by the use of bone morphogenetic protein-2 (BMP-2). The pull-out strengths of group A were greater than that of group B ($P < 0.05$). Scanning electronic microscopy (SEM) showed more calcified substances on the surface of the implants of group A than B. There was more marked bone around group A than B implants at 4 weeks ($P < 0.05$) and 8 weeks ($P < 0.05$). RhBMP-2 improves the quantity and quality of implant–bone osseointegration. Biomechanical testing and histomorphometric analysis are reliable methods to use in researching the implant–bone interface

12. **Wikesjö UM, Huang YH, Polimeni G, Qahash M.in 2007**[43] conducted a study to shown that rhBMP-2 induces normal physiologic bone in clinically relevant defects in the craniofacial skeleton. The newly formed bone assumes characteristics of the adjacent resident bone and allows placement, osseointegration /re-osseointegration, and functional loading of endosseous implants. Clinical studies optimizing dose, delivery technologies, and conditions for stimulation of bone growth will bring about a new era in dentistry. The ability to predictably promote osteogenesis through the use of BMP-technologies is not far from becoming a clinical reality and will undoubtedly have anastounding effect on how dentistry is practiced.

13. **Piemontese M, Aspriello SD, Rubini C, Ferrante L, Procaccini M. in 2008**[44] conducted a randomized, double-masked, clinical trial was to compare platelet-rich plasma (PRP) combined with a demineralized freeze-dried bone allograft (DFDBA) to DFDBA mixed with a saline solution in the treatment of human intrabony defects. Treatment with a combination of PRP and DFDBA led to a significantly greater clinical improvement in intrabony periodontal defects compared to DFDBA with saline. No statistically significant differences were observed in the hard tissue response between the two treatment groups, which confirmed that PRP had no effect on hard tissue fill or gain in new hard tissue formation.

14. **King GN, King N, Hughes FJ. in 2010**[45] conducted a study to investigate the effects of two different collagen delivery systems for rhBMP-2 in rat periodontal fenestration defects. Using the collagen membrane delivery system, 3 groups of adult Wistar rats which had surgical defects created on the right side of the mandible involving the removal of bone and exposure of the molar roots were treated with either rhBMP-2 in collagen membrane (BMPm) (n= 12 animals), or collagen membrane only (COLm) (n=12), or were left untreated (UN) (n= 14). Using the collagen gel delivery system, surgical defects were treated with either rhBMP-2 incorporated in a collagen gel carrier (BMPg) (n=5) or had collagen gel only (COLg) (n=6). In conclusion, both carrier systems for rhBMP-2 significantly increased new bone formation compared with controls during the early stages of periodontal wound healing. However, the more slowly dissolving collagen membrane carrier system for rhBMP-2 produced significantly greater new cementum compared with the collagen gel carrier, suggesting that a more prolonged exposure of rhBMP-2 is required to increase cementogenesis.

15. **Thoma DS, Jones A, Yamashita M, Edmunds R, Nevins M, Cochran DL.in 2010**[46] conducted a study on use of recombinant bone morphogenetic protein- 2 (rhBMP-2) with a collagen carrier material has severe limitations in regards to space maintenance. The aim of this study was to test whether rhBMP-2 combinations with allograft or a mesh enhance the regeneration of missing bone and the subsequent placement of dental implants. The combination of rhBMP-2 and a block allograft provides the greatest ridge width of the four treatment options used in this canine ridge augmentation model.

16. **Bashutski JD, Wang HL in 2011**[47] conducted a study on periodontal regeneration is preferred over tissue repair and is accomplished through the exclusion of epithelial tissues, which allows cementum, bone, and connective tissue to repopulate the wound. Recently, biologic materials have emerged as adjuncts to aid in regeneration by augmenting the events of wound healing in the area. A review of biologic agents was conducted using the following MeSH terms: guided tissue regeneration, intercellular signaling peptides and proteins, and biologic factors. Currently, EMD and PDGF have Food and Drug Administration approval for periodontal regeneration, whereas BMP-2 is approved for bone augmentation. FGF and PTH do not have Food and Drug Administration approval for periodontal applications and so their clinical usage is not indicated.

17. **Spagnoli DB, Marx RE 2011**[48] this article addresses the role of bone morphogenetic proteins (BMP) in native bone healing for implant attachments and the application of BMP to de novo bone regeneration associated with dental implants. The following two cases will illustrate the translation of a complex biology involving rhBMP-2/ACS, crushed cancellous freeze-dried allogeneic bone (CCFDAB), and platelet-rich plasma (PRP) into a predictable bone regeneration that provides a functional benefit to patients through the osseointegration of dental implants.

18. **Khojasteh A, Behnia H, Naghdi N, Esmaeelinejad M, Alikhassy Z, Stevens M in 2012**[49] reviewed the application and subsequent investigations in the use of varied osteogenic growth factors in bone regeneration procedures have grown dramatically over the past several years. Owing to this rapid gain in popularity and documentation, a review was undertaken to evaluate the in vivo effects of growth factors on bone regeneration. Within the limitations of this review, BMP-2 may be an appropriate growth factor for osteogenesis.

19. **Singh GR in 2013**[50] reviewed that BMPs have been tested in periodontal (regeneration of lost bone tissue due to periodontal disease), implant (increase in bone volume for placement of implants, maxillary sinus augmentation) and restorative-endodontic (pulpotomies) procedures.

20. **Hur BM and Lim SB in 2014[51]** conducted a clinical trial that aims to evaluate the effect of rhBMP-2 compared with bioactive glass on the periodontal tissue regeneration in 2- and 3-wall intrabony periodontal defect. 23 patients(male 13 and female 10) who had probing depth above 5 mm in one wall and the more of tooth walls received Biogran® bone grafts in 14 control sites and CowellBMP® in 13 experimental sites. The probing depth and gingival recession were measured at the baseline; 3 month and 6 months after surgery Biogran® and CowellBMP® were effective in treatment of infra-bony periodontal defects. CowellBMP® was more significantly effective in decrease of probing depth and the increase of probing attachment level than Biogran®.

21. **Chadwick JK, Mills MP, Mealey BL in 2016[52]** conducted a study on wide variety of materials have been proposed for treatment of periodontal intrabony bony defects; recently, platelet-rich fibrin has been suggested as a grafting material. The aim of this study is to report changes in clinical attachment level and bone fill of periodontal intrabony defects treated with demineralized freeze dried bone allograft (DFDBA) compared to platelet-rich fibrin (PRF) in humans. Treatment of intrabony defects with either DFDBA or PRF resulted in a significant gain in CAL as well as bone fill after 6 months of healing, with no significant difference between materials.

22. **Jaiswal Y, Kumar S, Mishra V, Bansal P, Anand KR, Singh S in 2017[53]** conducted a study to access the efficacy of decalcified freeze-dried bone allograft (DFDBA) in the regeneration of bone following small osseous defect in minor oral surgery. Twenty patients with cysts were assessed. Ten patients were filled with DFDBA (Group 1) and ten without bone graft (Group 2), respectively. Radiographic bone density was assessed on preoperative, intraoperative, and postoperative radiographs on 1st day, 3rd month, and at 6th month using Adobe Photoshop CS6 - Grayscale histogram. Bone formed as depicted by bone density is significantly higher when DFDBA is used in small bony defects.

23. **Schorn L, Sproll C, Ommerborn M, Naujoks C, Kübler NR, Depprich R in 2017[54]** conducted a study focuses on the three dimensional vertical bone generation in a one stage procedure in vivo. Therefore, a collagenous disc-shaped scaffold (ICBM = Insoluble Collagenous Bone Matrix) containing rhBMP-2 (Bone Morphogenetic

Protein-2) and/or VEGF (Vascular Endothelial Growth Factor) was applied around the coronal part of a dental implant during insertion. RhBMP-2 and VEGF released directly at the implantation site were assumed to induce the generation of new vertical bone around the implant By using collagenous disc-shaped matrices in combination with rhBMP-2 and VEGF vertical bone can be generated in a one stage procedure without donor site morbidity. The results of the presenting study suggest that the combination of rhBMP-2 and VEGF applied locally by using a collagenous carrier improves vertical bone generation in vivo. Further research is needed to establish whether this technique is applicable in clinical routines.

24. **Bavsar AK, Prabhuji ML, Varadhan KB, Parween in 2018**[55] reviewed regeneration is reproduction or reconstitution of injured or lost part with the growth and differentiation of new cells and intercellular substances to form new tissues or parts. Periodontal regeneration refers to healing after periodontal surgery that result in the restoration of the lost periodontium and attachment apparatus viz. cementum, alveolar bone and periodontal ligament. Treatment of periodontal disease has evolved from just fighting bacteria to a combined effort to eliminate the offending microorganisms, to arrest the progression of tissue damage and to regenerate lost tissues. Although some of the regenerative techniques have been available for several years, and some have shown promising results, none of the techniques are without problems and none have proven to be 100% effective. In perspective, periodontal regeneration remains a challenging and complex endeavour, requiring synchronous formation of all periodontal tissues via cementogenesis, osteogenesis and formation of a periodontal ligament, generating a similar form and function found in the intact, native periodontal attachment.

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25. **Petsos H et al in 2019**[56] conducted a study in originally 16 periodontitis patients (baseline examination) periodontal surgery was performed in 44 infrabony defects. Polylactide acetyltributyl citrate barriers were randomly assigned to 23 out of these 44 defects (parallel). Ten of these patients (GTR) exhibited a second, contra-lateral defect (OFD) each (split-mouth). At baseline, 12, 120 and 240□12 months after surgery probing depths, attachment level, bleeding on probing as well was Plaque Index, Gingival Bleeding Index and Plaque Control Record were obtained Twenty years after OFD and GTR in infrabony defects in a population with lack of regular

SPT attachment gains at 12 months after surgery were stable. 82% of the initially included teeth were still in place.

26. **Alhussaini AH in 2019**[57] conducted a study on two bioactive materials were compared to evaluate their effect on dental implant stability. A total of 32 patients (102 dental implants) were divided into 3 groups: 24 dental implants with bone morphogenetic protein (BMP), 27 dental implants with PRF, and 51 dental implants without BMP or PRF (control group). Data were statistically analyzed to determine the bioactive material with the best effect on implant stability. Dental implants coated with BMP have a better effect on stability than those with PRF alone and those without PRF or BMP.

27. **Atchuta A, Gooty JR, Guntakandla VR, Palakuru SK, Durvasula S, Palaparthy R. in 2020**[58] conducted a study on several bone graft materials are popularized in the treatment of intrabony defects. Demineralized freeze-dried bone allograft (DFDBA) is widely used in the treatment of intrabony defects. Platelet rich fibrin (PRF) is autologous blood preparation which helps in wound healing and regeneration. Hence, this study focuses on evaluation of PRF, DFDBA, and their combination in the regeneration of intrabony defects. Combination of DFDBA and PRF improved the clinical and radiographic parameters compared to PRF and DFDBA alone. PRF was combined with DFDBA to produce a synergistic effect for treating intrabony defects in chronic periodontitis patients.

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MATERIALS AND METHODOLOGY

Materials And Methodology

Place of the study where it is conducted:-

This clinical, experimental prospective study was carried out in the Department of Periodontology, Babu Banarasi Das College of Dental Sciences (BBDCODS), Babu Banarsi Das University (BBDU) Lucknow.

Study Sample and size

- Group A- 10 Patients were treated with Demineralized Freeze-Dried Bone Allografts (DFDBA) alone.
- Group B- 10 Patients were treated with Demineralized Freeze- Dried Bone Allografts (DFDBA) + Recombinant Human Bone Morphogenetic Protein -2 (rhBMP-2).
- ***Inclusion criteria:***

1. Patients in the age group of 35-60 years.
2. Patients suffering from Chronic Periodontitis with Probing Pocket depth ≥ 6mm.
3. Patients with radiographic evidence of intrabony defects.
4. Patients fulfilling ASA Physical status classification system criteria.

- ***Exclusion Criteria:***

1. Patients with any systemic diseases that affects the periodontal treatment outcome.
2. Pregnant and lactating women.
3. Smokers and tobacco chewers.
4. Patients who have used antibiotics for the previous 3 months.
5. Subjects with a known allergy to the material being used.
6. Non co-operative patients.

ARMAMENTARIUM AND MATERIALS

- Surgical gloves face masks, head cap, face shield and suction tip.
- Sterile cotton and gauge.
- Normal saline, povidone iodine solution and 0.2% chlorhexidine gluconate solution.
- Mouth mirrors, UNC-15 Probe (HuFriedy®), Tweezers and Explorer.
- Syringe 3ml and 5ml.
- Local anaesthetic agent 2% Lignocaine Hydrochloride and Adrenaline bitartrate (1:80000).
- BP blade handle, Blade No. 12, 15C and 15, Periosteal elevator (HuFriedy®, P24).
- A set of Gracey curettes (HuFriedy®) and Columbia curettes (HuFriedy®).
- Cumin scaler (HuFriedy®).
- Bone graft carrier and condenser (GDC).
- Adson tissue holding forceps (GDC).
- Castroviejo scissors (GDC) and needle holder (GDC).
- Demineralized freeze – dried bone allograft (DFDBA) [Tata Memorial Hospital Mumbai].
- Recombinant Bone Morphogenetic Protein-2 (rhBMP-2) [COWELL® BMP Cowellmedi Co., ltd. The Pioneers in dental Implant & E.rhbmp-2 Busan, Republic of Korea]
- Mersilk sutures-(4-0) [Braided Silk Black] (ETHICON).
- COE-PAK™ Periodontal dressing. (GC AMERICA INC.).

CLINICAL PARAMETERS

All the clinical parameters (PI, GI, PPD and CAL) and radiographic parameters were recorded at the baseline (after scaling and root planing) and after 6 months.

- **Plaque Index (Silness and Loe,)[59]**

The Plaque Index (PI) is fundamentally based on the same principle as the Gingival Index, namely the desirability of distinguishing clearly between the severity and the location of the soft debris aggregates. The purpose of introducing this system (Silness

and Löe, 1964) was also to create a plaque index which would match the Gingival Index completely.

Criteria for the plaque index system

0 = No plaque in the gingival area.

1 = A film of plaque adhering to the free gingival margin and adjacent area of the tooth. The plaque may only be recognized by running a probe across the tooth surface.

2 = Moderate accumulation of soft deposits within the gingival pocket, on the gingival margin and/or adjacent tooth surface, which can be seen by the naked eye.

3 = Abundance of soft matter within the gingival pocket and/or on the gingival margin and adjacent tooth surface.

Each of the four gingival areas of the tooth is given a score from 0-3; this is the PI for the area. The scores from the four areas of the tooth may be added and divided by four to give the PI for the tooth. The scores for individual teeth (incisors, premolars and molars) may be grouped to designate the PI for the groups of teeth. Finally, by adding the indices for the teeth and dividing by the number of teeth examined, the PI for the individual is obtained.

PI I = 0 is the score given when the gingival area of the tooth surface is literally free of plaque.

PI I = 1 represents the situation where the gingival area is covered with a thin film of plaque which is not visible, but which is made visible.

PI I = 2 is the score given when the deposit is visible in situ

P1I = 3 is reserved for the heavy (1-2 mm. thick) accumulation of soft matter.

- **Gingival Index (Loe and Silness)[59]**

The gingival index (GI), a tool for evaluating the intensity and scope of gingival inflammation in both individuals and subjects within sizable demographic groupings, was first proposed in 1963. The GI just evaluates the gingival tissues. Each of the four gingival regions of the tooth—the face, mesial, distal, and lingual—is examined for inflammation using this procedure, and the degree of inflammation is quantified by assigning each area a score between 0 and 3. A periodontal probe is used to examine bleeding by moving it over the gingival crevice's soft tissue wall. To determine the

tooth score, add the scores for the four tooth locations and divide the result by 4.By adding the tooth scores together and dividing by the number of teeth examined, an individual's GI score can be obtained.

Scores and Criteria for Gingival Index (GI)

0 = Normal gingiva.

1 = Mild inflammation: slight change in color and slight edema; no bleeding on probing.

2 = Moderate inflammation: redness, edema, and glazing; bleeding on probing.

3=Severe inflammation: marked redness and edema; ulceration; tendency to spontaneous bleeding.

- **Probing pocket depth[60]**

The probe-able crevice's bottom is measured from the gingival edge in order to determine the depth of the probe (i.e., where the probe tip stops).

Exploration with a periodontal probe is the only reliable way to locate and measure periodontal pockets. By using a radiographic examination, pockets are not found. An alteration to soft tissue is the periodontal pocket. Radiographs show areas of bone loss where pockets may be suspected, but they do not show the presence or depth of pockets, therefore they do not distinguish between the presence of pockets before and after their removal unless the bone has been altered.

In cases of gingival inflammation, probing depth is often greater than 3 mm and less than 3 mm in cases of gingival health. Numerous investigations have been conducted to establish the probe's depth of penetration in a pocket or sulcus. Beagle dogs were employed by Armitage and colleagues8 to assess the probe's penetration when a standard force of 25 g was applied.

- **Clinical Attachment Level[61]**

The term "attachment level" refers to the region on a tooth where the dentogingival junction first appears coronally. The distance between the attachment level and a

reference point on a tooth, like the cementoenamel junction, is measured by clinical attachment level. Gains or losses in attachment can cause changes in the attachment level, which can give a more accurate indicator of the level of periodontal gain or destruction.

Clinical attachment loss (CAL) is used to categorise the severity of chronic periodontitis into three categories: mild (1–2 mm CAL), moderate (3–4 mm CAL), and severe (>5 mm CAL).

- **Radiographic evaluation:**

  Bone level was measured with the help of ImageJ™ software. It is a Java-based image processing program developed at the National Institutes of Health and the Laboratory for Optical and Computational (LOCI, University of Wisconsin). After taking the IOPAR, bone level was measured from CEJ to the deepest point in the intrabony defect for both the groups first at the baseline then after 6 months.

SURGICAL PROCEDURE:

All the clinical and radiographic parameters were recorded at baseline. After the recordings, the patient was asked for a pre-procedural rinse with 10 ml of 0.2% chlorhexidine gluconate solution for 60 seconds. The surgical procedures were performed under aseptic conditions. The operative site was anesthetized with a solution of 2% lignocaine with 1:80000 adrenaline. Sulcular incisions were given with the help of 15C and 12 no. BP blade and full thickness flap was reflected with the help of periodontal elevator (Hufriedy® P24). The calculus removal and root planing were done in intrabony defect, after the removal of granulation tissue with the help of Gracey, Columbia and Universal curettes (Hufriedy®). The tissue tags were removed using Castroveijo scissors.

Surgical area was irrigated with povidone iodine and was carefully inspected to ensure the complete debridement of granulation tissue. The defect site in Group A was grafted with demineralized freeze- dried bone allograft (DFDBA) and in Group B the defect site was grafted with DFDBA and rhBMP-2(COWELL ® BMP). The graft was mixed with normal saline and was placed into the defect. Condensation of the graft was done by using bone graft condenser and care was taken to avoid the overfilling of the defect

so as to ensure an adequate closure of the flap. Also over-condensation was avoided for sufficient vascularization within the graft and to prevent any infection. The flap was sutured in close approximation using interrupted sutures (4-0) [Mersilk, ETHICON]. Surgical site was protected by applying a periodontal dressing (COE-PAK™ Periodontal dressing. GC AMERICA INC.).

Amoxicillin 500 mg TDS and Acelophenac 100 mg in combination with paracetamol 325 mg BD were prescribed for both the groups for 5 days. Patient was recalled after 10 days for sutures and dressing removal. Plaque control was reinforced at the time of suture removal. Further recalls for clinical and radiographic re-evaluation were schedule at 6 months. At each visit, plaque control measures will be reinforced and supra gingival scaling will be done if required.

SURGICAL ARMAMENTARIUM

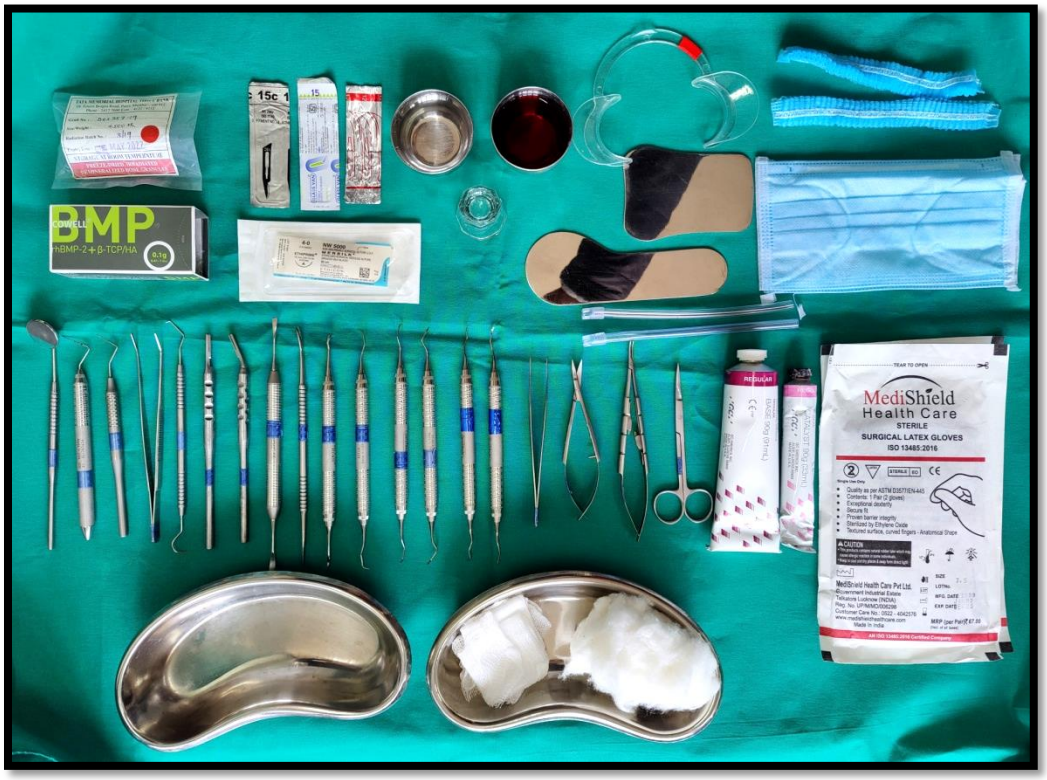


Figure 1: Armamentarium

Figure 2: DFDBA and RhBMP-2

GROUP-1 DFDBA

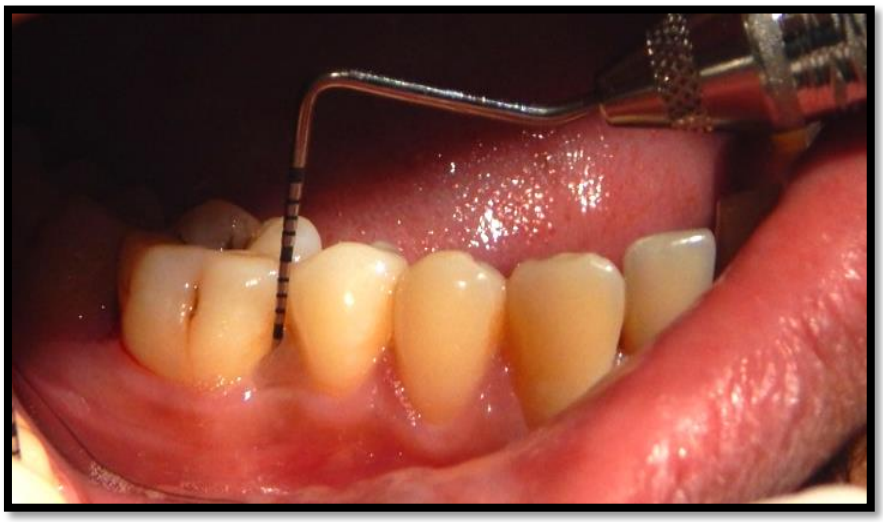


Figure 3(i): Pre-Operative Probing Pocket Depth (Buccal)

Figure 3(ii): Pre-Operative Probing Pocket Depth (Lingual)

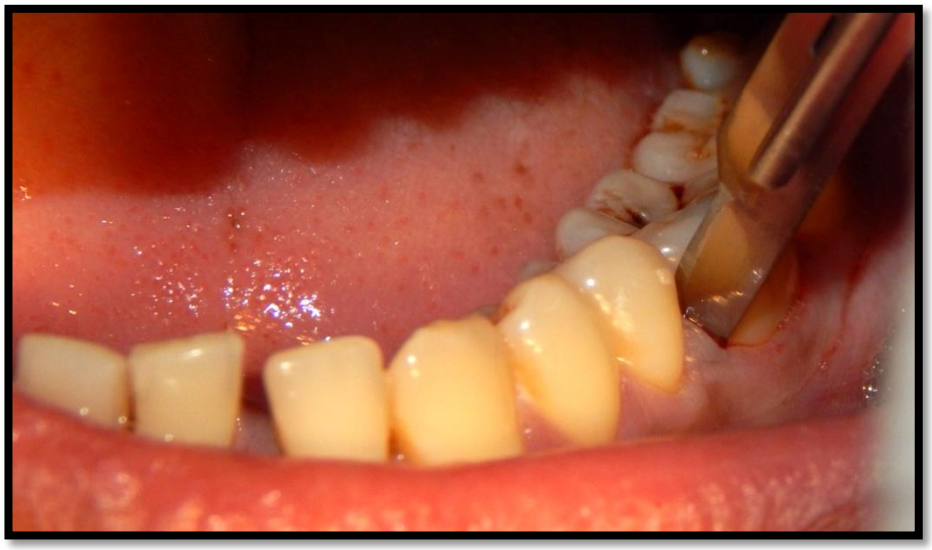


Figure 3(iii): Crevicular Incision

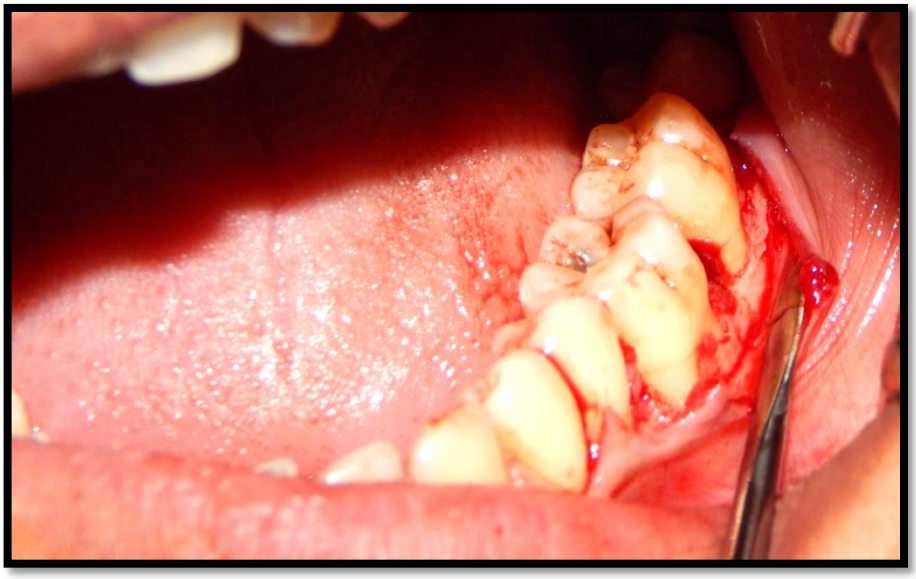


Figure 3(iv): Post Debridement

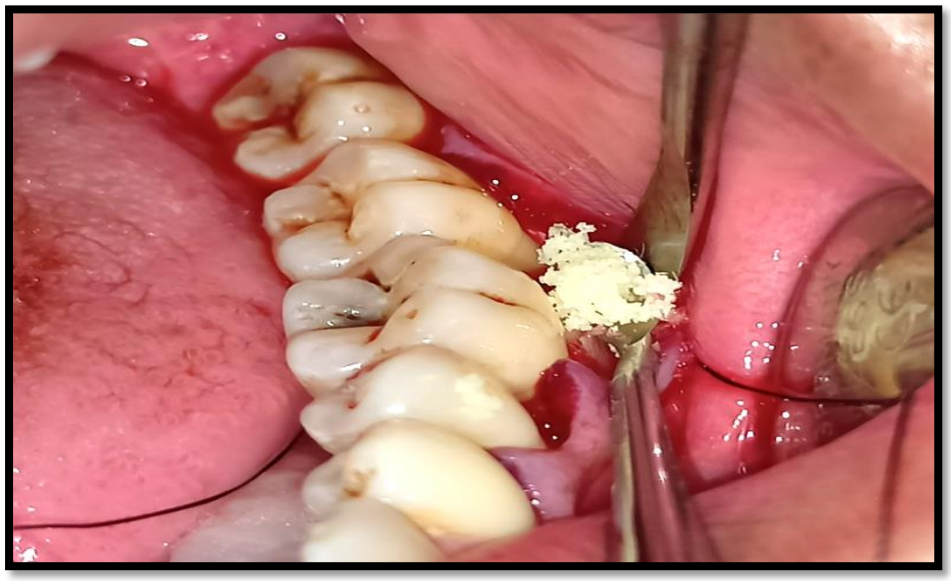


Figure 3(v): Placement of DFDBA in the defect

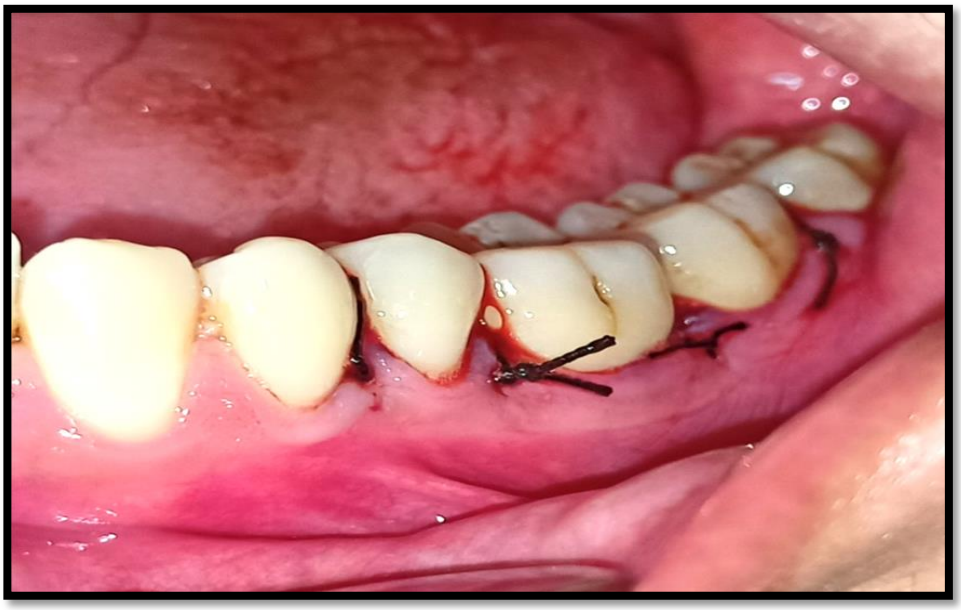


Figure 3(vi): Flap approximated with Sutures

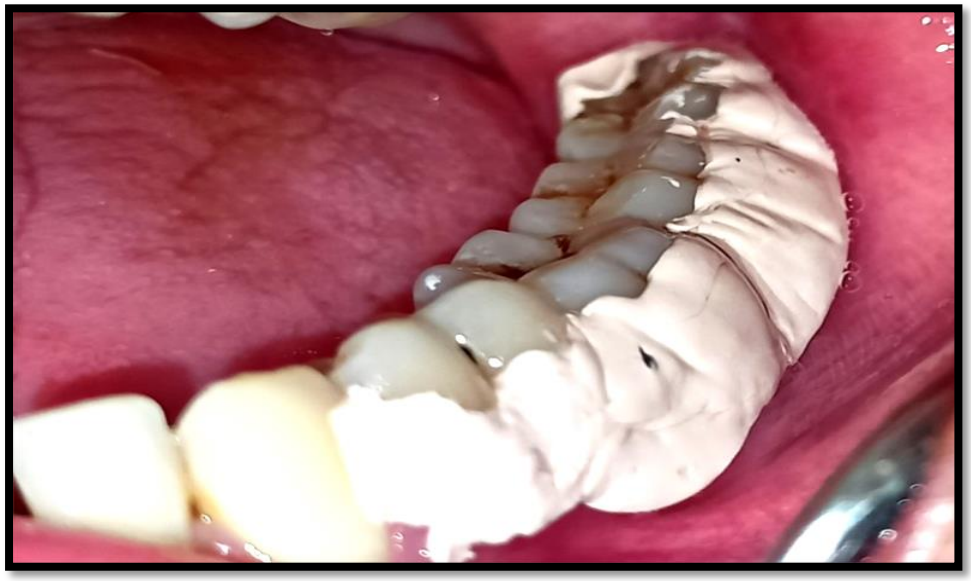


Figure 3(vii): Periodontal Dressing

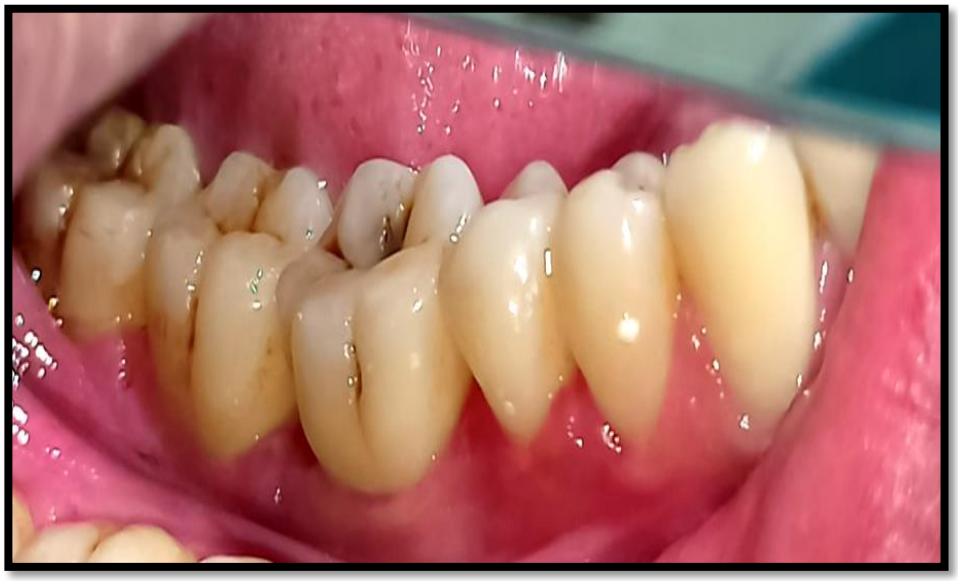


Figure 3(viii): Suture removal after 14 days

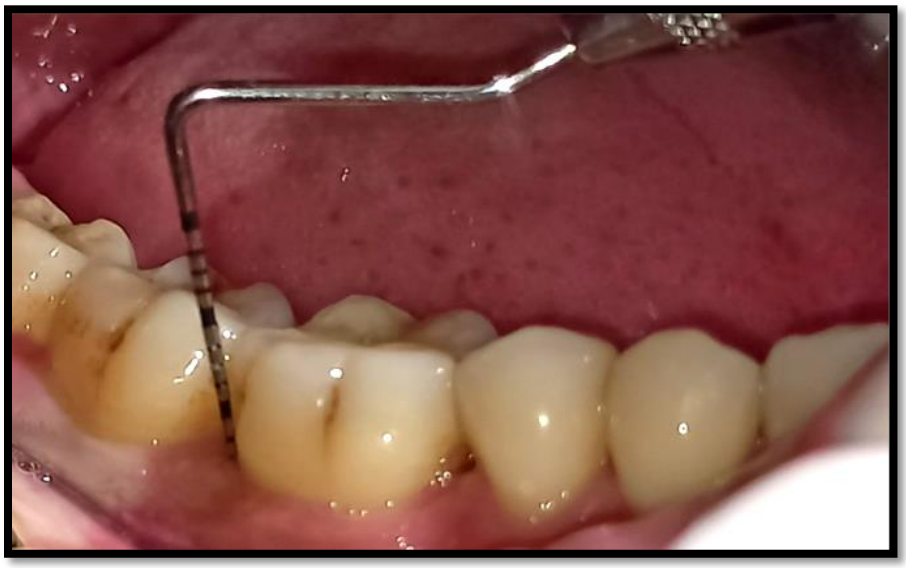


Figure 3(ix): Post Operative Probing Pocket Depth

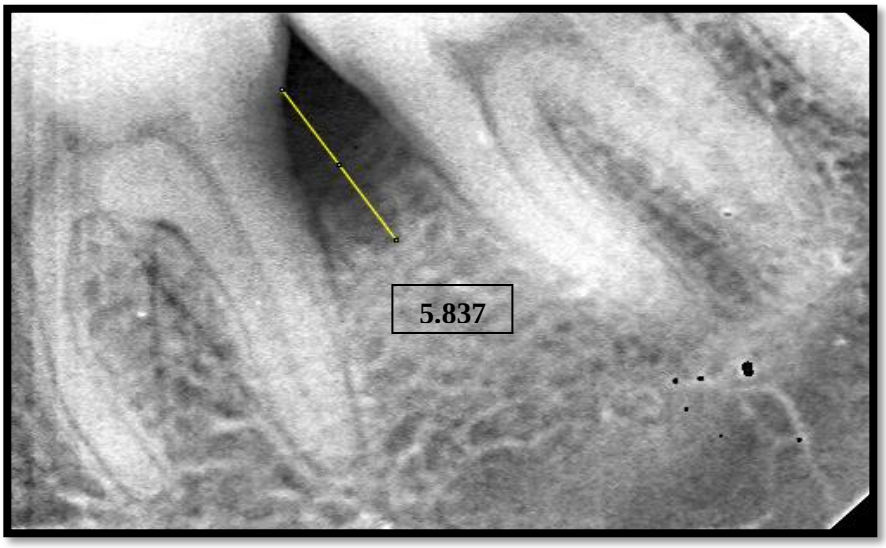


Figure 3(x): IOPAR at Baseline

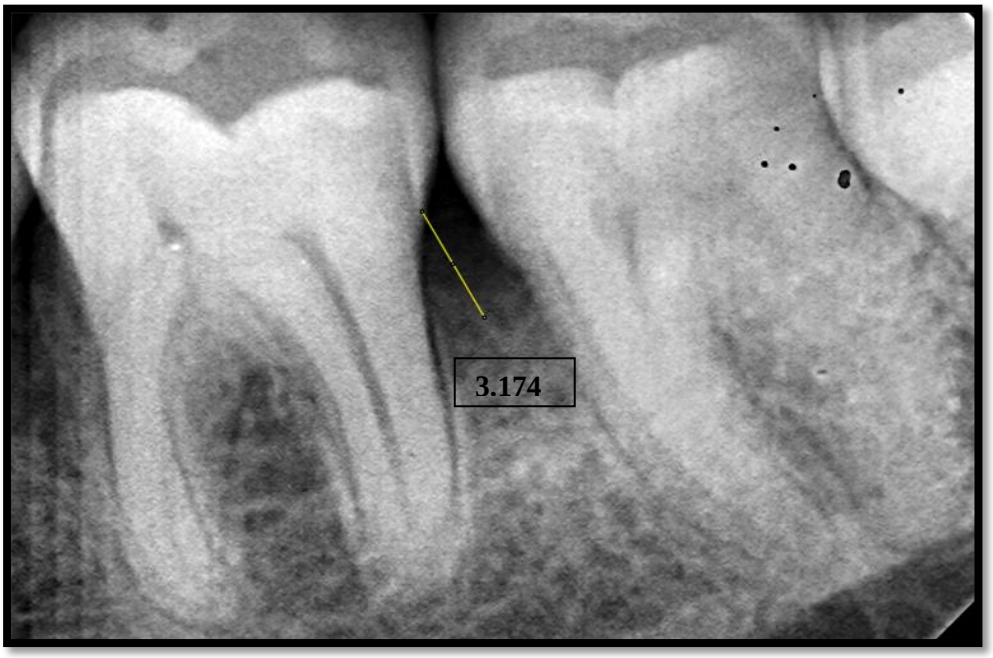


Figure 3(xi): IOPAR at 6months

GROUP-2 DFDBA with RhBMP-2

Figure 4(i): Pre-Operative Probing Pocket Depth (Buccal)

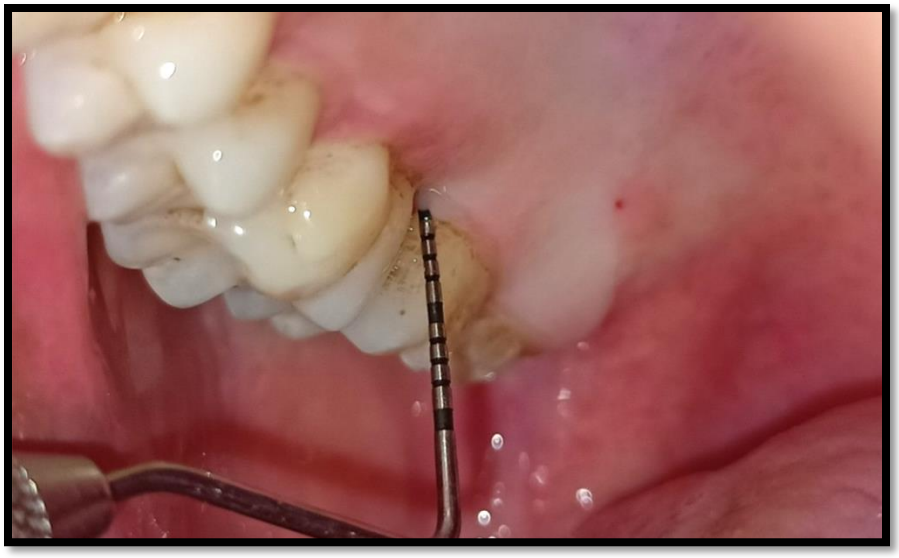


Figure 4(ii): Pre-Operative Probing Pocket Depth (Palatal)

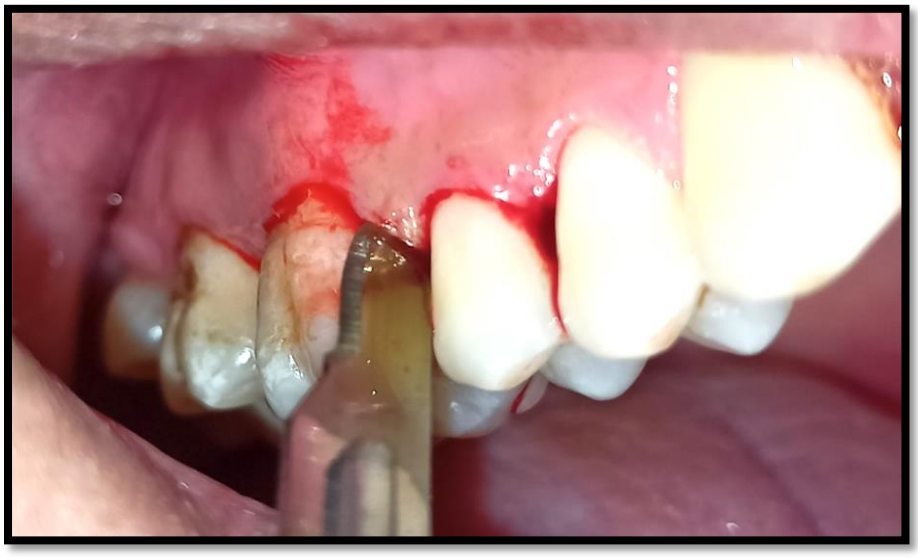


Figure 4(iii): Crevicular Incision

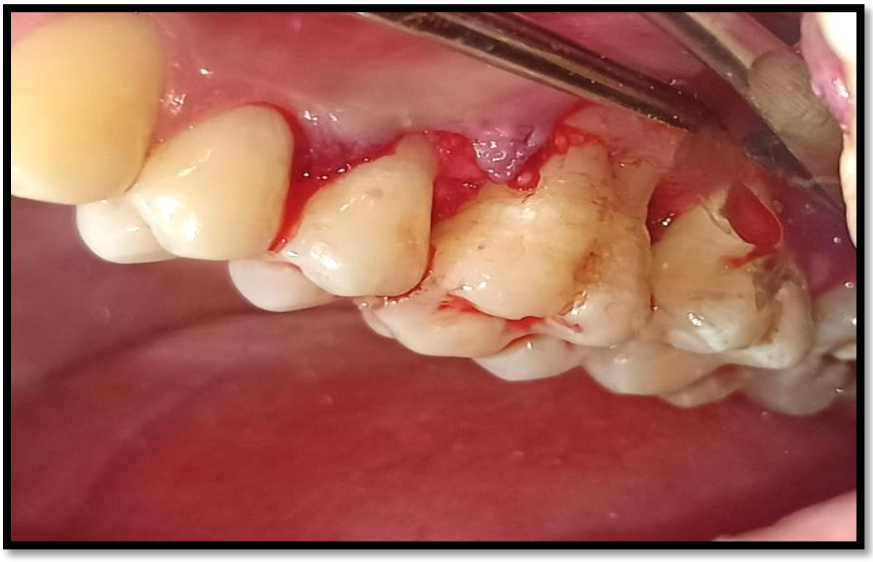


Figure 4(iv): Post Debridement

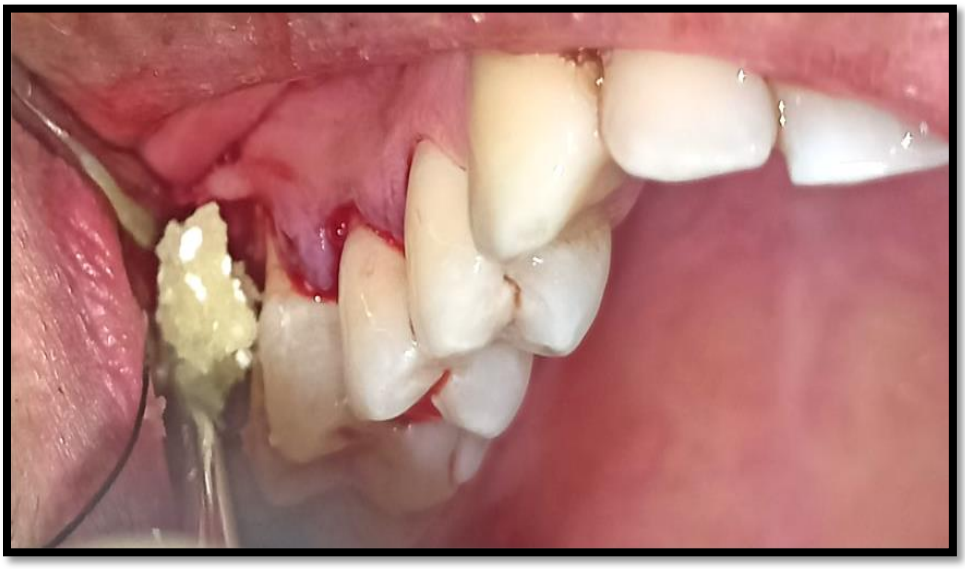


Figure 4(v): Placement of DFDBA and rhBMP-2 in the defect

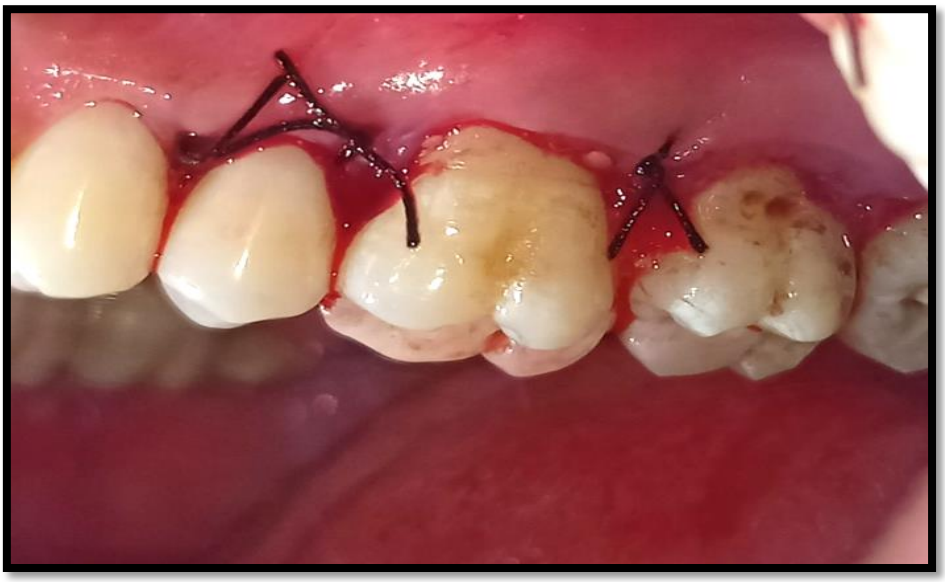


Figure 4(vi): Flap approximated with Sutures

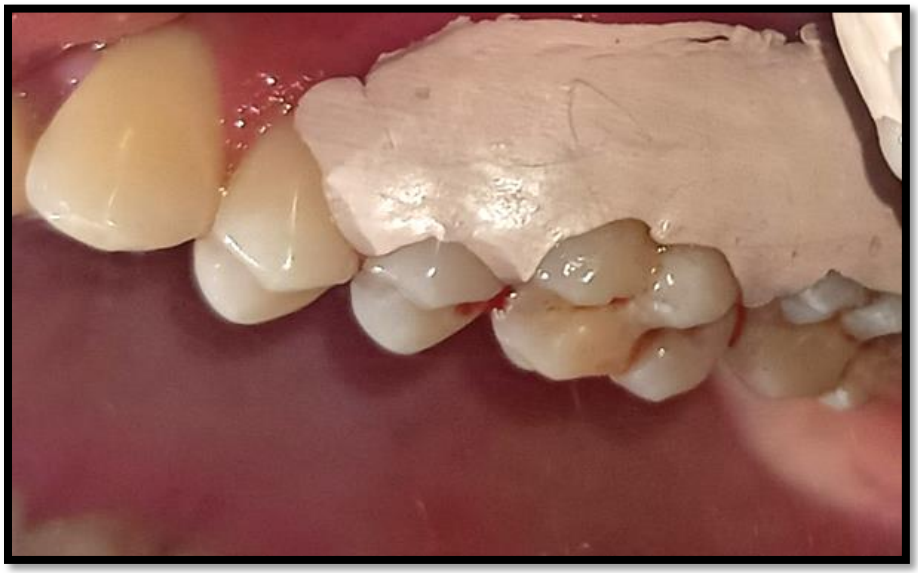


Figure 4(vii): Periodontal Dressing

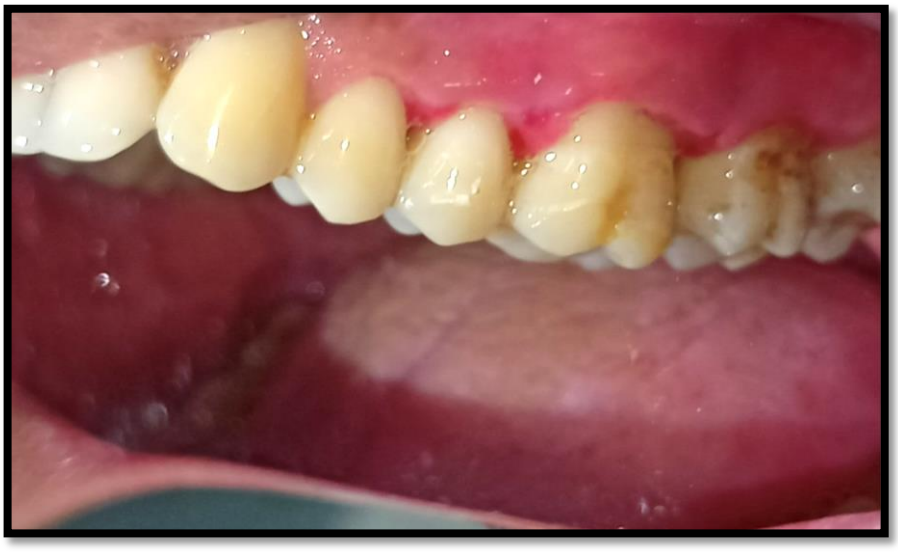


Figure 4(viii): Suture removal after 14 days

Figure 4(ix): Post Operative Probing Pocket Depth

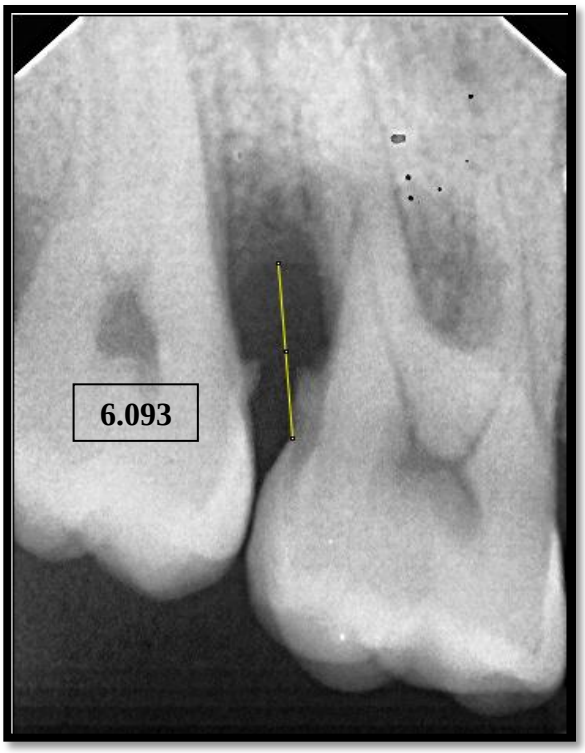


Figure 4(x): IOPAR at Baseline

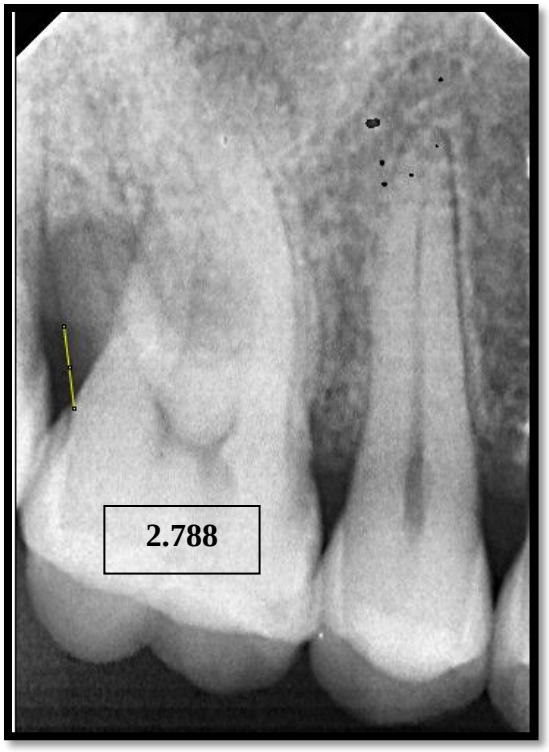


Figure 4(xi): IOPAR at 6months

OBSERVATIONS AND RESULTS

OBSERVATIONS AND RESULTS

Section 1: Comparative assessment between Group A and Group B (Inter Group)

Table 1: Comparative evaluation of Plaque – Baseline between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	2.5000	.52705	.00000	.23570	.000	1.000
Group B	10	2.5000	.52705				

P-value < 0.05

10 patients each in Group A and Group B were evaluated for periodontal variables. Plaque index at baseline was 2.500 ± 0.52705 for both Group A and Group B patients. Thus having no significant difference between them at p=1.000 as seen in Table 1 and Graph 1.

Graph 1: Comparative evaluation of Plaque – Baseline between groups

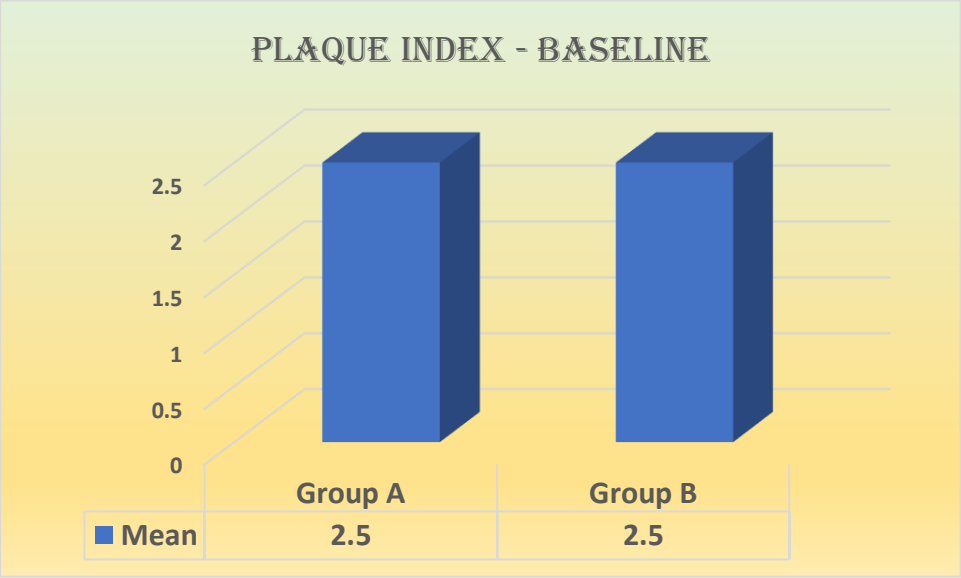


Table 2: Comparative evaluation of Plaque at 6 months between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	1.0000	.00000[a]	-	-	-	-
Group B	10	1.0000	.00000[a]				

P-value < 0.05

Plaque Index after 6 months of intervention reduced to 1.000 + 0.000 in both groups as seen in Table 2 and Graph 2.

Graph 2: Comparative evaluation of Plaque at 6 months between groups

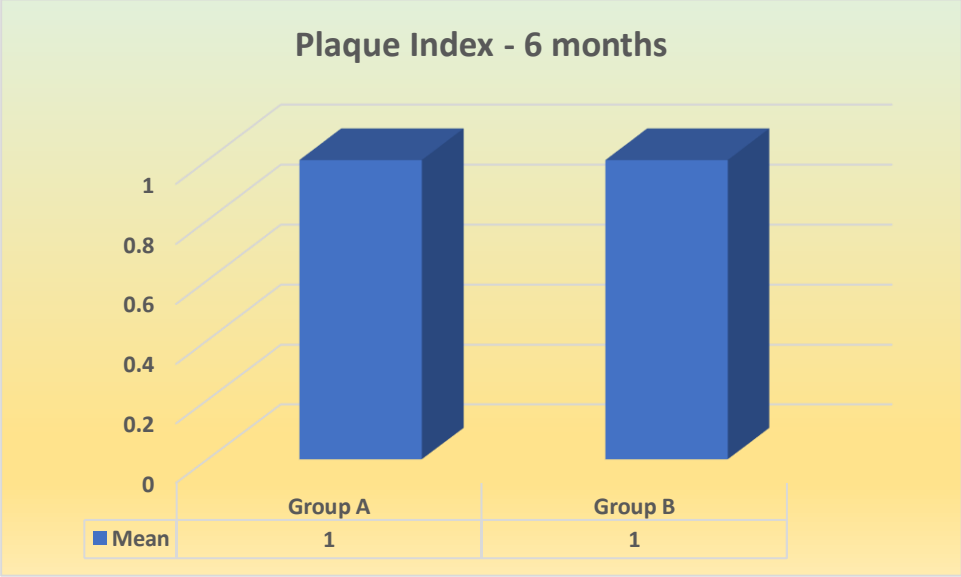


Table 3: Comparative evaluation of Gingival Index at baseline between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	2.5800	.24404	.08000	.11813	.677	.507
Group B	10	2.5000	.28284				

P-value < 0.05

Gingival Index at baseline between the groups were almost similar in both groups, non significant at p=0.507 as seen in Table 3 and Graph 3.

Graph 3: Comparative evaluation of Gingival Index at baseline between groups

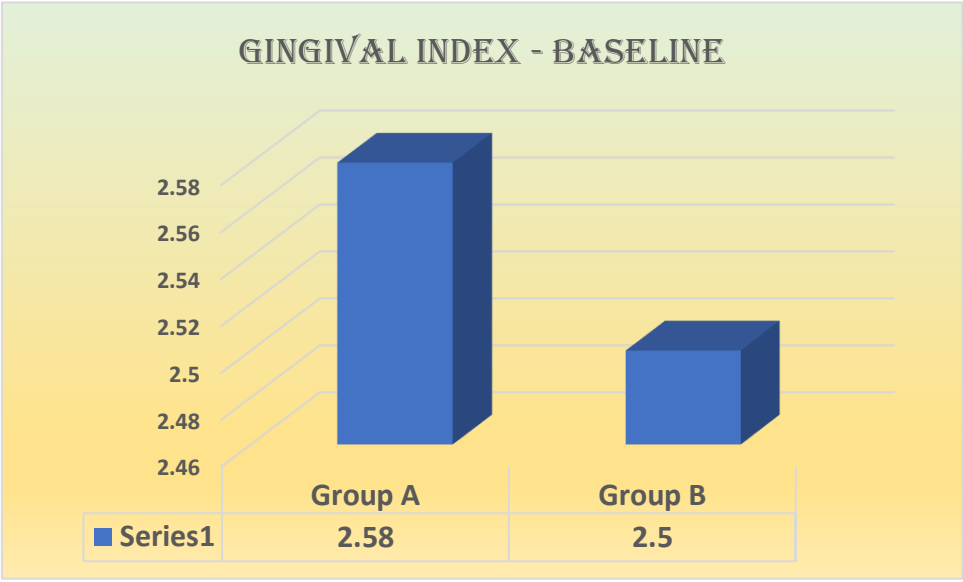


Table 4: Comparative evaluation of Gingival Index at 6 months between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	.5700	.26268	.10000	.11935	.838	.413
Group B	10	.4700	.27101				

P-value < 0.05

Group A had a GI of .5700 ± .2626 and Group B had .4700 ± .27101, after 6 months which was not significant.

Graph 4: Comparative evaluation of Gingival Index at 6 months between groups

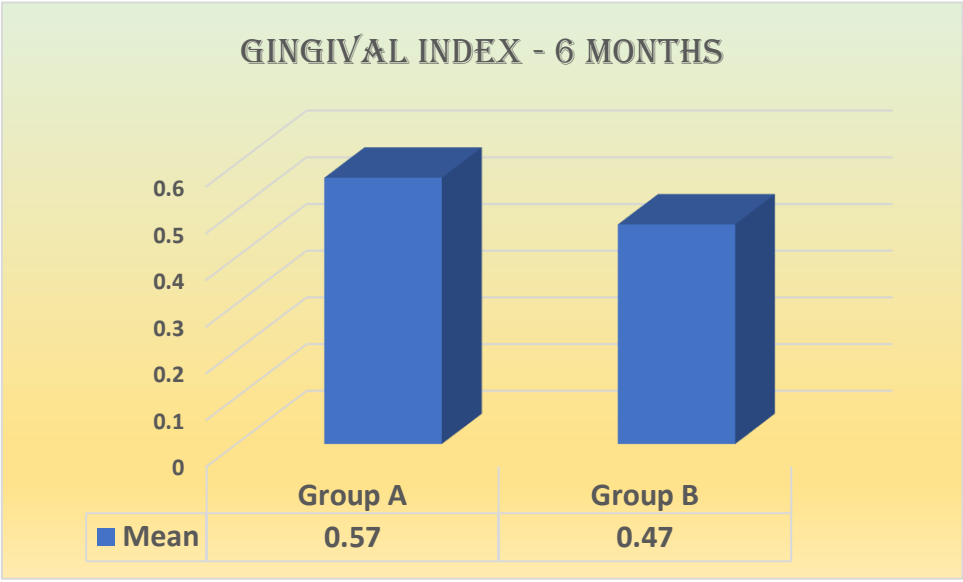


Table 5: Comparative evaluation of PPD at baseline between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	7.0000	.94281	.00000	.39441	.000	1.000
Group B	10	7.0000	.81650				

P-value < 0.05

Probing pocket depth at baseline was similar in both group at 7.000 ± 0.9428and 7.000 ±0.81650, non significant at p=1.00 as observed in Table 5 and Graph 5.

Graph 5: Comparative evaluation of PPD at baseline between groups

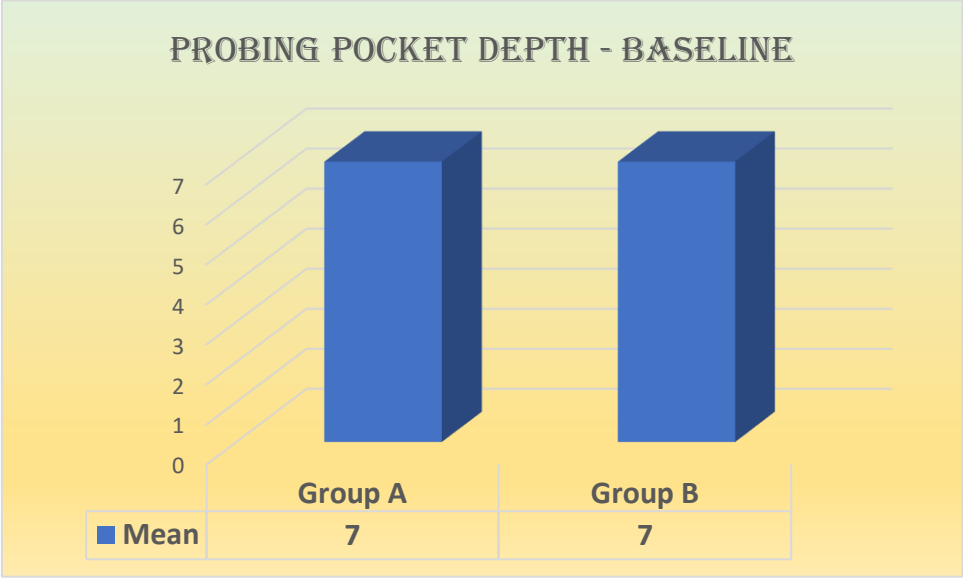


Table 6: Comparative evaluation of PPD at 6 months between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	3.4000	.69921	.00000	.31269	.000	1.000
Group B	10	3.4000	.69921				

P-value < 0.05

At the end of six months, reduction in PPD in both groups was at par with each other for Group A and Group B, as shown in Table 6 and Graph 6.

Graph 6: Comparative evaluation of PPD at 6 months between groups

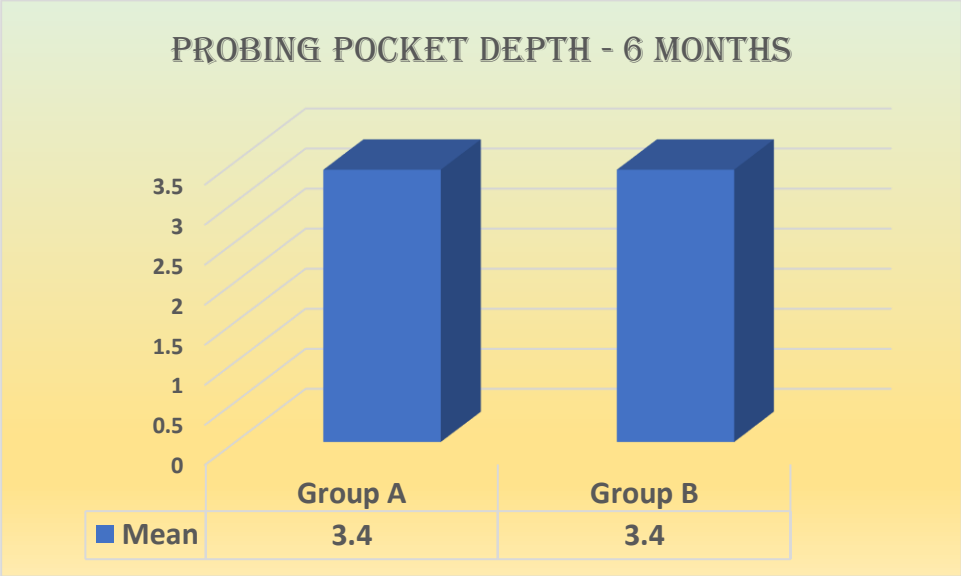


Table 7: Comparative evaluation of CAL at baseline between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	6.50	.84984	-.20000	.37417	. -.535	.600
Group B	10	6.7000	.82327				

P-value < 0.05

Clinical attachment level at base line scored similar in both Group A and Group B as seen in Table 7 and Graph 7.

Graph 7: Comparative evaluation of CAL at baseline between groups

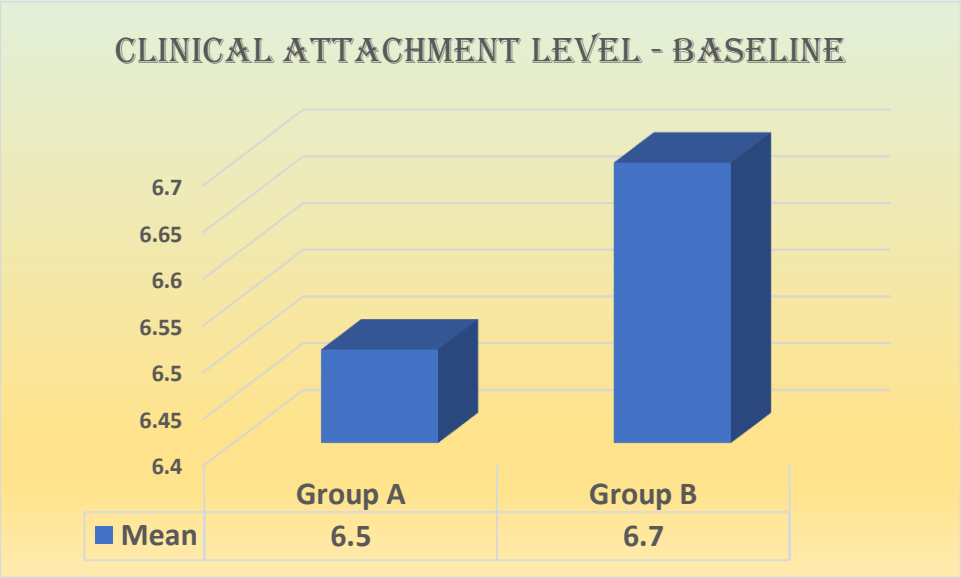


Table 8: Comparative evaluation of CAL at 6 months between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	2.6000	.84327	-.70000	.37268	-1.878	.077
Group B	10	3.3000	.82327				

P-value < 0.05

When compared between the groups Group A performed slightly better with CAL scoring to 2.600 +.84327 as against 3.3000 + .82327 in Group B, but the difference was not significant as seen in Table 8 and Graph 8.

Graph 8: Comparative evaluation of CAL at 6 months between groups

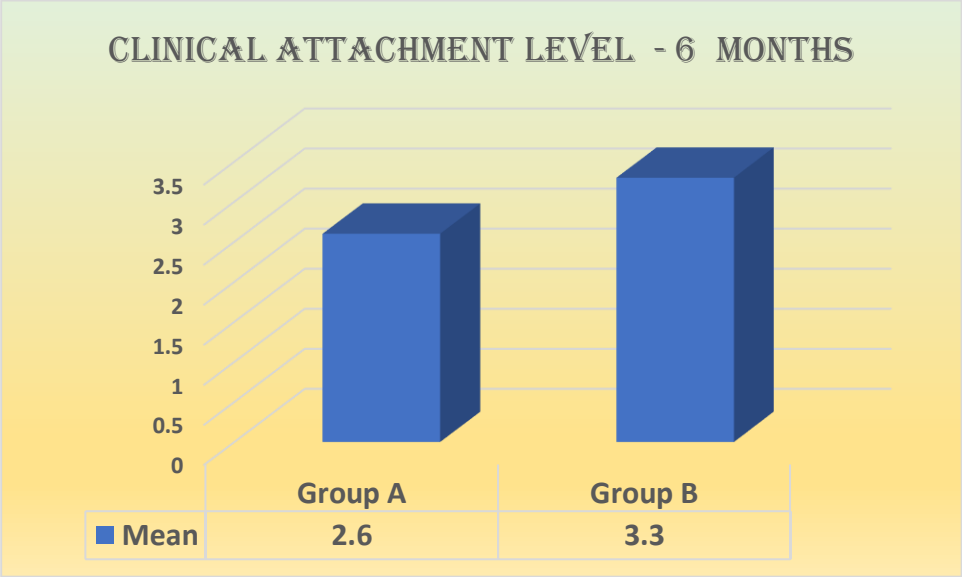


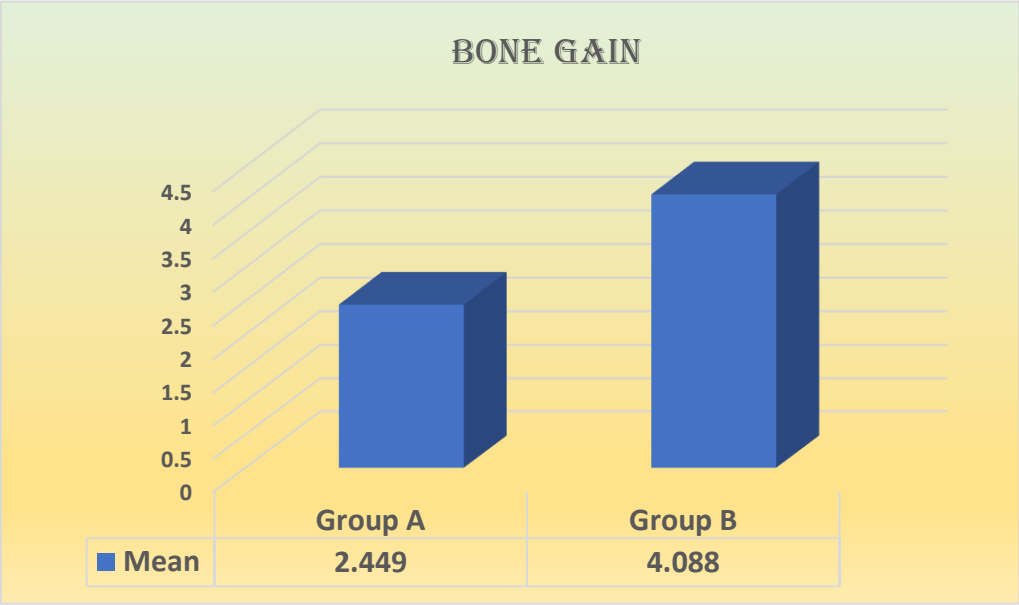
Table 9: Comparative evaluation of Bone gain between groups

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Group A	10	2.4490	.34719	-1.63900	.16774	-9.771	0.000
Group B	10	4.0880	.40105				

P-value < 0.05

When assessed for bone gain between groups, Group A showed bone gain of 2.4490 ± .34719 while it was 4.0880 ± .40105 in the Group B which was statistically significant at p=0.000 as demonstrated in Table 9 and graph 9.

Graph 9: Comparative evaluation of Bone gain between groups



Section 2: Comparative assessment in Group A – Baseline and 6 months (Intra Group)

Table 10: Comparative evaluation of Plaque Index in Group A

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	2.5000	.52705	1.50000	.52705	9.000	0.000
6 months	10	1.0000	.00000				

P-value < 0.05

Baseline and 6 months post operative assessment of Plaque Index in Group A showed a significant reduction of 1.5000, significant at p = 0.000.

Graph 10: Comparative evaluation of Plaque Index in Group A

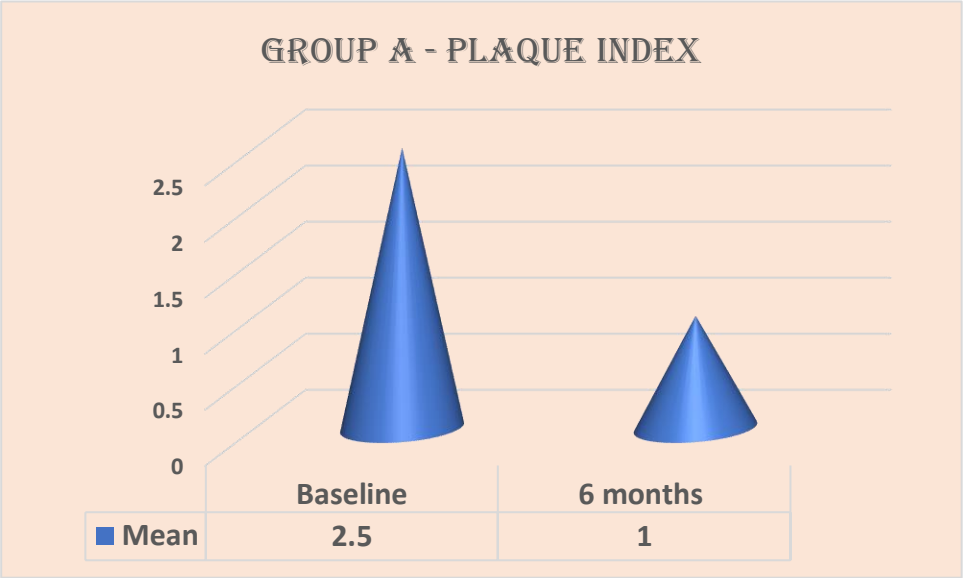


Table 11: Comparative evaluation of Gingival Index in Group A

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	2.5800	.24404	2.01000	.32128	19.784	0.000
6 months	10	.5700	.26268				

P-value < 0.05

Gingival Index significantly reduced in Group A subjects from 2.5800 ±.24404 to .5700 ± .26268 from baseline to 6 months of intervention.

Graph 11: Comparative evaluation of Gingival Index in Group A

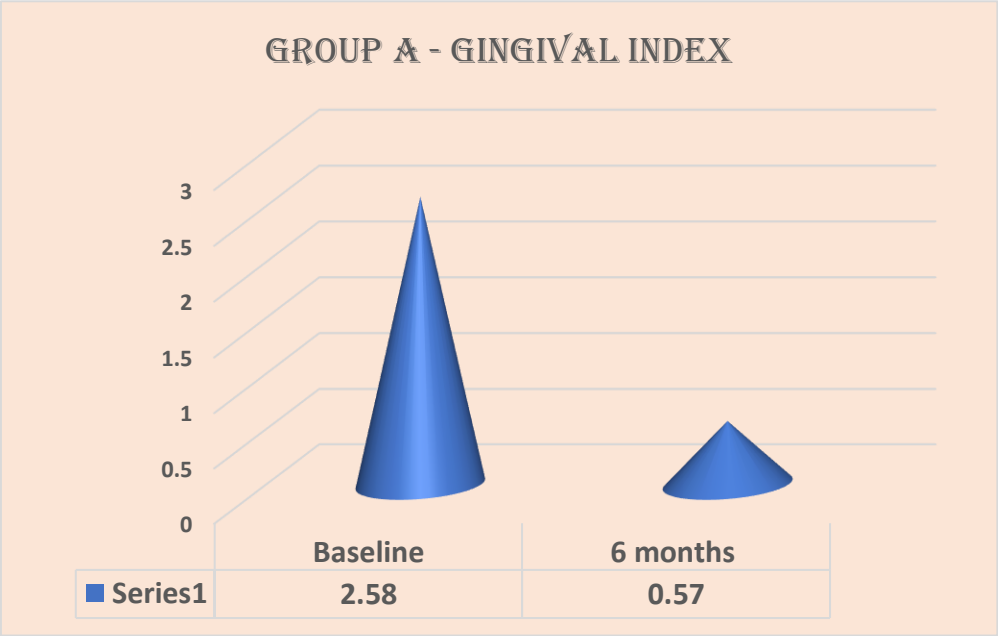


Table 12: Comparative evaluation of Periodontal Pocket depth in Group A

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	7.0000	.94281	3.60000	.51640	22.045	0.000
6 months	10	3.4000	.69921				

P-value < 0.05

Periodontal pocket depth exhibited significant reduction from baseline score of 7.000 ± .94231 to 3.4000 ± .69921 after 6 months, significant at p = 0.000 in Group A participants.

Graph 12: Comparative evaluation of Periodontal Pocket depth in Group A

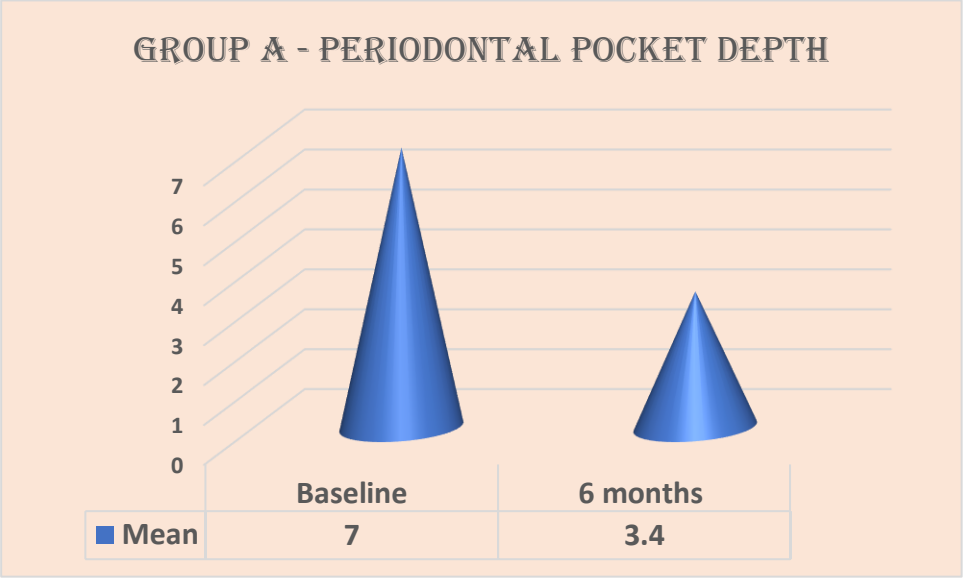


Table 13: Comparative evaluation of Clinical Attachment Level in Group A

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	6.5000	.84984	3.90000	.73786	16.714	0.000
6 months	10	2.6000	.84327				

P-value < 0.05

A significant reduction was noted in Clinical attachment level in group A from 6.5000 ± .84984 to 2.6000 ± .84327 at 6 months of intervention which was significant at p=0.000 as observed in Table 13 and Graph 13.

Graph 13: Comparative evaluation of Clinical Attachment Level in Group A

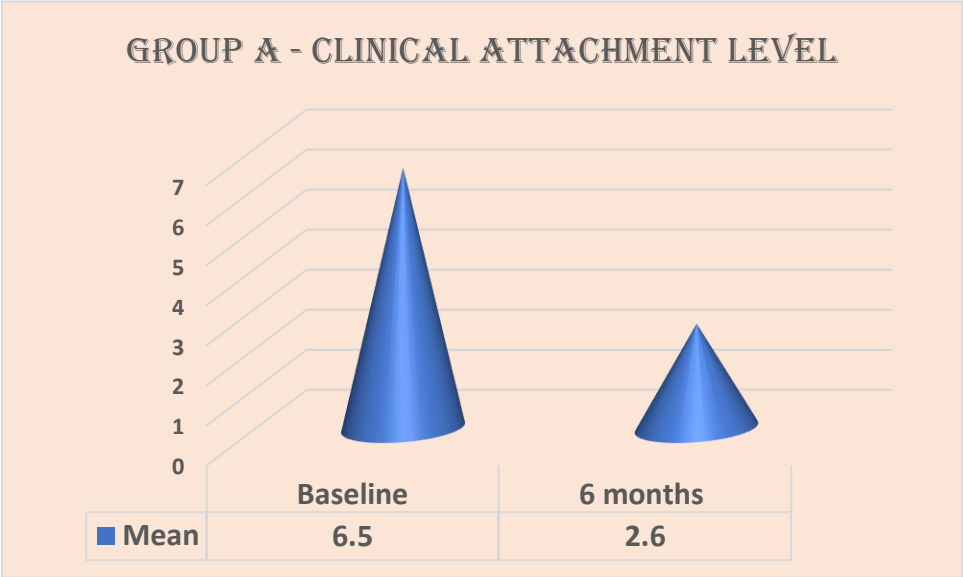


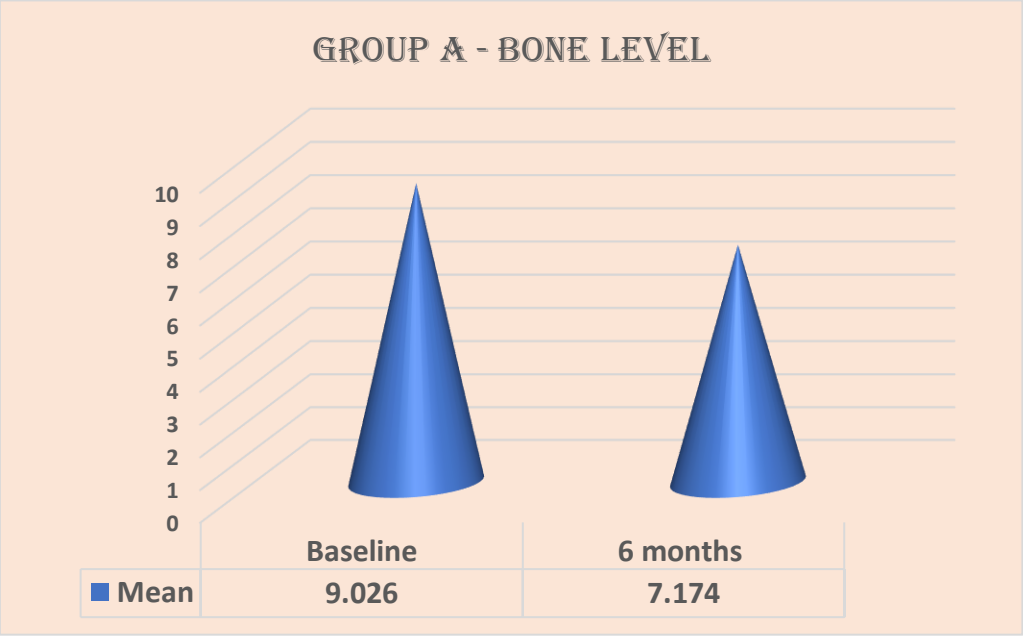
Table 14: Comparative evaluation of Bone level in Group A

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	9.0260	1.68670	1.85200	.45740	12.804	0.000
6 months	10	7.1740	1.74510				

P-value < 0.05

A significant reduction was noted in Bone level in group A from 9.0260 ± 1.68670 to 7.1740 ± 1.74510 at 6 months of intervention which was significant at p=0.000 as observed in Table 14 and Graph 14

Graph 14: Comparative evaluation of Bone level in Group A



Section 3: Comparative assessment in Group B – Baseline and 6 months (Intra Group)

Table 15: Comparative evaluation of Plaque Index in Group B

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	2.5000	.52705	1.50000	.52705	9.000	0.000
6 months	10	1.0000	.00000				

P-value < 0.05

Baseline and 6 months post operative assessment of Plaque Index in Group B showed a significant reduction of 1.5000, significant at p = 0.000.

Graph 15: Comparative evaluation of Plaque Index in Group B

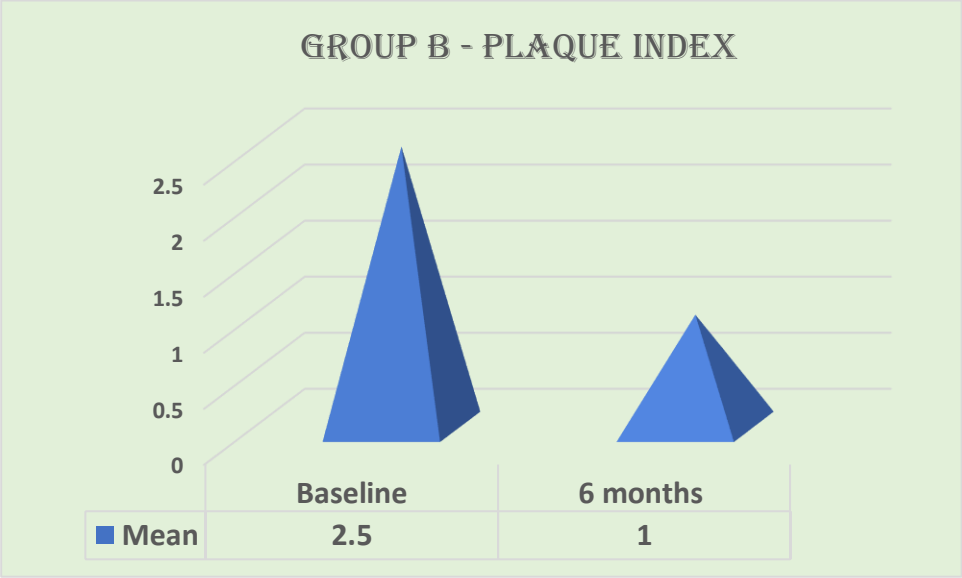


Table 16: Comparative evaluation of Gingival Index in Group B

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	2.5000	.28284	2.03000	.08233	77.974	0.000
6 months	10	.4700	.27101				

P-value < 0.05

Gingival Index significantly reduced in Group B subjects from 2.500 ±.28284 to .4700 ± .27101 from baseline to 6 months of intervention.

Graph 16: Comparative evaluation of Gingival Index in Group B

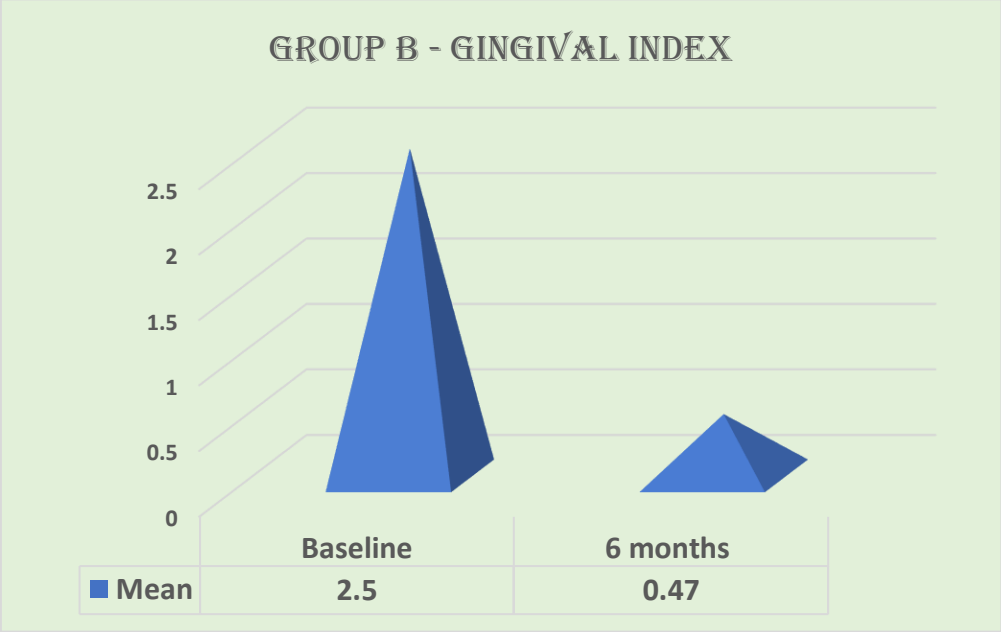


Table 17: Comparative evaluation of Periodontal Pocket depth in Group B

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	7.0000	.81650	3.60000	.51640	22.045	0.000
6 months	10	3.4000	.69921				

P-value < 0.05

Periodontal pocket depth exhibited significant reduction from baseline score of 7.000 ± .81650 to 3.4000 ± .69921 after 6 months, significant at p = 0.000 in Group B participants.

Graph 17: Comparative evaluation of Periodontal Pocket depth in Group B

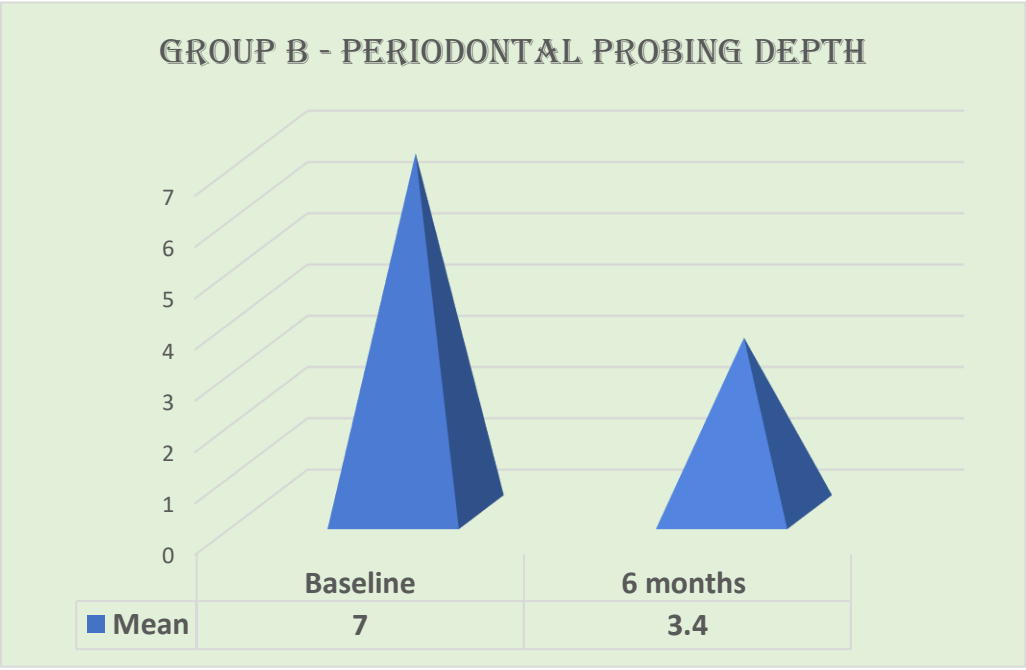


Table 18: Comparative evaluation of Clinical Attachment Level in Group B

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	6.7000	.82327	3.40000	.51640	20.821	0.000
6 months	10	3.3000	.82327				

P-value < 0.05

A significant reduction was noted in Clinical attachment level in group B from 6.7000 ± .82327 to 3.3000 ± .82327 at 6 months of intervention which was significant at p=0.000 as observed in Table 18 and Graph 18.

Graph 18: Comparative evaluation of Clinical Attachment Level in Group B

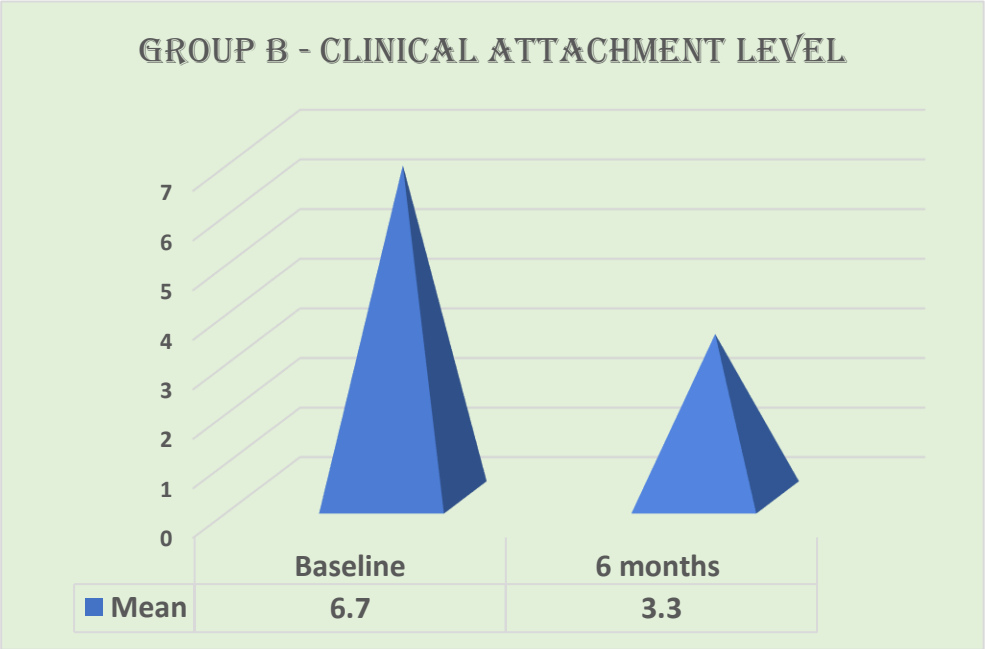


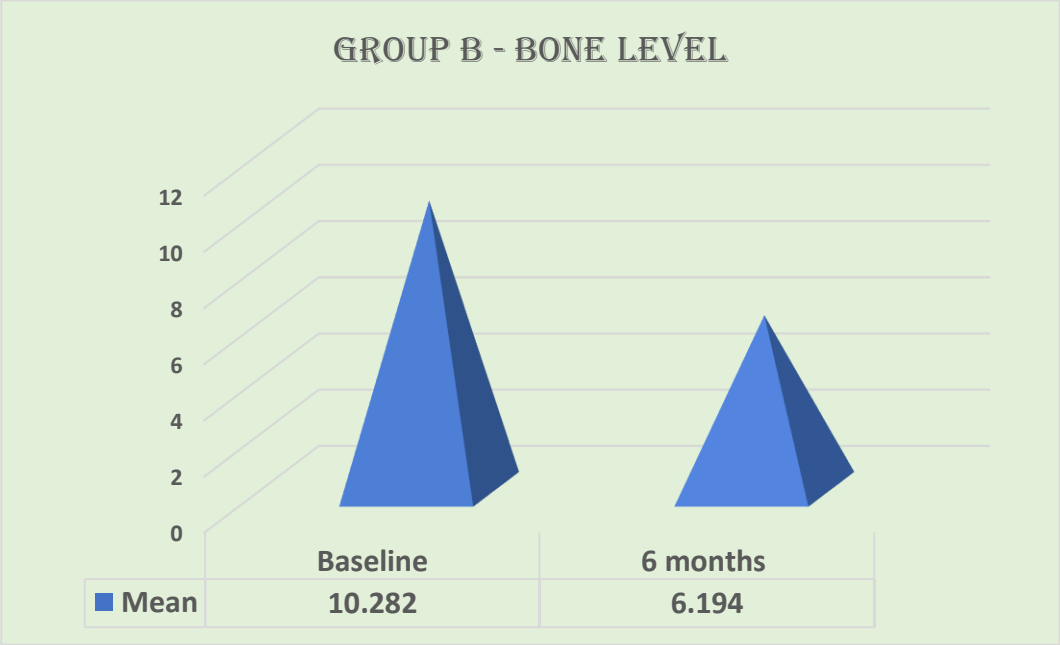
Table 19: Comparative evaluation of Bone level in Group B

Groups	N	Mean	St. Deviation	Mean Difference	Std. Error Difference	't' statistic	P value
Baseline	10	10.2820	.75867	4.08800	.40105	32.234	0.000
6 months	10	6.1940	.64163				

P-value < 0.05

A significant reduction was noted in Bone level in group A from 10.2820 ± .75867 to 6.1940 ± .64163 at 6 months of intervention which was significant at p=0.000 as observed in Table 19 and Graph 19

Graph 19: Comparative evaluation of Bone level in Group B



DISCUSSION

DISCUSSION

The ultimate goal of periodontal therapy is the creation of an environment that is conducive to maintain patient dentition in a state of optimum health, comfort and function. The deep intraosseous periodontal defects presents a major challenge in achieving the goal as it increases the risk of disease progression and recurrence after systematic traditional periodontal therapy. Regenerative periodontal therapy aims to reform and reconstitute the supporting tissues of teeth which have been lost due to periodontal disease and trauma. Several regenerative therapeutic procedures have been developed for this purpose; have met with partial or marginal success. These include root surface biomodification, use of various types of bone grafts, guided tissue regeneration and combination of the above.

There have been numerous therapeutic grafting modalities investigated for restoring periodontal osseous defects. It is critical to understand the following bone graft material classifications: **Autografts** are bone from the same individual; **allografts** are bone from a different individual of the same species; and **xenografts** are bone from a different species.

Bone graft materials are examined for their osteogenic, osteoinductive, osteoconductive or osteopromotion capabilities.

Ellegaard *et al*. (1973, 1974, 1975, 1976) and Nielsen *et al*. (1980, 1981) reported that grafting materials in periodontal bony defects may be:[62-65]

Osteoproliferative (osteogenetic): new bone is formed by bone-forming cells contained in the grafted material.

Osteoconductive: the grafted material does not contribute to new bone formation *per se* but serves as a scaffold for bone formation originating from adjacent host bone.

Osteoinductive: bone formation is induced in the surrounding soft tissue immediately adjacent to the grafted material.

When the grafted material lacks osteoinductive qualities but nonetheless increases osteoinduction by encouraging bone development, this is known as **osteopromotion**. As an illustration, it has been demonstrated that while enamel matrix derivatives alone

do not promote de novo bone development, when combined with demineralized freeze-dried bone allograft (DFDBA), they improve DFDBA's osteoinductive properties.[66]

Autogenous Bone Grafts

Autogenous bone grafts, also called autografts, are bone grafts transferred from one site to another site within the same individual. These grafts are the **gold standard** to which all other grafting materials are compared. Autogenous grafts can be cortical or cancellous or a combination of both.[67]

- Bone from Intraoral Sites

Hegedüs attempted to use bone grafts to reconstruct bone defects caused by periodontal disease in 1923[68]. Nabers and O'Leary revived the method in 1965[69], and numerous efforts have been made since then to define its indications and technique.

Bone from healing extraction wounds, edentulous ridges, bone trephined from within the jaw without damaging the roots, newly formed bone in wounds specially created for the purpose, bone removed from tuberosity and the ramus, and bone removed during osteoplasty and ostectomy are all sources of bone[70].

Osseous Coagulum: Robinson described a technique using a mixture of bone dust and blood that he termed “osseous coagulum.” The technique uses small particles ground from cortical bone. The advantage of the smaller particle size is that it provides additional surface area for the interaction of cellular and vascular elements.[71]

Bone blend: The bone blend technique uses an autoclaved plastic capsule and pestle. Bone is removed from a predetermined site, triturated in the capsule to a workable, plastic like mass, and packed into bony defects.[72]

Cancellous bone can be obtained from the maxillary tuberosity, edentulous areas, and healing sockets. Cancellous bone and marrow are removed with curettes, back-action chisels, or trephine.

The bone swaging technique requires an edentulous area adjacent to the defect, from which the bone is pushed into contact with the root surface without fracturing the bone at its base. Bone swaging is technically difficult, and its usefulness is limited.[73]

- Bone from Extraoral Sites.

Hegedüs also pioneered the use of bone from the tibia as a source of bone for grafting into periodontal osseous defects in 1923[68]. In the 1960s, Schallhorn and Hiatt revived this approach by using the iliac crest.

Extraoral bone graft harvesting is a common technique, particularly when many bone grafts are needed. According to many writers, extraoral cancellous bone and marrow grafts have the best chance of generating new bone.[74-76] The iliac crest is the best location to obtain extraoral bone grafts. Since they have been demonstrated to promote cementogenesis, bone regeneration, and Sharpey's fibre reattachment, autografts from iliac cancellous bone and marrow have a significant osteogenic potential, according to Rosen et al. (2000).[77] Postoperative problems, cost, time, and the need for a further surgical treatment are all significant drawbacks of extraoral iliac grafts. Autogenous iliac crest transplants are a less ideal alternative due to all of these concerns.

Allografts.

A graft obtained from genetically different members of the same species is known as an allograft. Allografts are harvested from fresh cadavers under sterile conditions, typically within 24 hours of the donor's passing. Their unrestricted availability and osteoinductive capacity that rivals autogenous bone are their main advantages. Freeze-dried bone allograft (FDBA) and decalcified freeze- dried bone allograft are the two main types that are offered (DFDBA).

By exposing bone morphogenic proteins and other inductive factors that are known to promote bone formation, demineralization of the cortical bone allograft enhances its osteoinductive capacity. For this reason, while DFDBA additionally offers an osteoinductive surface in addition to an osteoconductive scaffold, FDBA only provides an osteoconductive scaffold and induces resorption when implanted in mesenchymal tissues. Allografts must be obtained, processed, and sterilised in accordance with established criteria (FDBA/DFDBA). According to the Centers for Disease Control and Prevention, the majority of bone banks follow the AATB's (American Association of Tissue Banks) regulations.[78]

According to AATB, allografts should not be collected if,

- Medical evaluations and behavioural risk assessments have revealed that the donor belongs to high-risk categories.

- The ELISA test results for the donor's HIV antibodies were positive.
- Donor autopsy reveals occult illness.
- A test for bacterial contamination of the donor's bone came up positive.
- The hepatitis B surface antigen (HBSAG) or hepatitis C virus was detected in the donor and bone (HCV).
- Syphilis testing on the donor was positive.

Processing of allografts:

The following describes the fundamental procedure of bone processing, even if allograft manufacturing businesses do not publicly publish the precise technology they use.

Obtaining bone from a competent donor and cutting it into pieces that are no more than 5 mm in size is the first and most crucial step. The second step is the elimination of bone marrow and cellular waste. Bone marrow and cellular waste are removed using liquids and detergents, which improves the bone's capacity to conduct osteoconductive energy. Pressure allows chemicals that inactivate or eliminate bacteria to fully penetrate the bone. To get rid of bioburden and reduce antigenicity, this technique uses chemical solutions such as saline, acetone, ethanol, or hydrogen peroxide. Then, bone fragments are treated with antibacterial, antimycotic, and antifungal treatments. After that, bone pieces are kept in liquid nitrogen at -80°C, a very low temperature. The bone fragments are then freeze-dried. Logistically, freeze-drying is advantageous since it prolongs the time that the tissue may be kept at room temperature. Bone fragments are regularly cleansed using solvents to remove moisture content. The size of the bone fragments is then further decreased to between 250 and 750 m. After the FDBA processing is complete, the graft is subsequently put into sterile containers. The graft is then placed in low-temperature, low-dose y radiation to guarantee sterility.

DFDBA is then processed through the decalcification process after reaching a final particle size of 250 to 750 m. The bone pieces are immersed in a hydrochloric acid bath with a pH range of 0.5 to 0.6 N for varied lengths of time. These acid-treated particles are then immersed in a buffering solution to eliminate any leftover acid. The demineralized allograft is subsequently washed with various solvents to remove any leftover buffer solution (such distilled water). The graft is then enclosed in sterile

containers and subjected to low-dose gamma radiation at low temperatures to ensure sterility.

After bone transplant processing, an exponential drop in graft contamination and/or disease transmission is observed. The likelihood that an item won't be sterile after going through a validated sterilisation process is known as the "sterility assurance level" (SAL).[79] A SAL of 10 is frequently attained by allografts for dental use with suitable processing. In other words, the likelihood of a bacterium surviving following allograft processing is less than one in a million.[80]Following processing, bone allograft must pass several tests, including:

Visual inspection test: Visual detection is used to spot problems including extensive graft contamination, flawed packaging, and mislabeled goods.

Residual moisture test: To confirm that the residual moisture is 6 percent or below, FDBA is tested.

Residual calcium test**:** To make sure that the residual calcium concentration is 8% or less, DFDBA is tested.

The allograft is packaged and sent for clinical use once it has passed all of the aforementioned tests.

Initial research by Urist[13], and others showed that DFDBA has the ability to induce osteoinduction and started to clarify the processes involved in DFDBA's induction of mineralization both in vitro and in vivo. These studies and later examinations of the mineralized matrix of bone's composition have demonstrated that a number of important noncollagenous proteins play an important role in bone's ability to promote bone growth. These comprise the bone morphogenetic proteins [BMPs] and members of the transforming growth factor ß superfamily (TGFßs), in addition to additional growth factors and adhesion molecules. Importantly, we still don't fully comprehend the mechanisms and circumstances required to induce biomineralization, despite the fact that evidence to date suggests that BMPs can promote the formation of minerals.

Most likely, other noncollagenous proteins in addition to the BMPs are required for formation of "functional" bone and may include osteocalcin (bone "gla" protein),

matrix "gla" protein, osteonectin, osteopontin (OPN), bone sialoprotein (BSP), bone acidic glycoprotein- 75 (BAG-75), thrombospondin, proteoglycans, and serum proteins.

Based on these findings, DFDBA ought to be a fantastic source of the osteoinductive elements needed to encourage biomineralization.

The main findings regarding inductive capacity were, first, that particle size did not correspond with inductive capacity, despite the fact that DFDBA preparations differ in both particle size and ability to stimulate new bone formation. Second, there was a wide range in how effective DFDBA batches were at promoting bone growth, including the observation that two batches from the same supplier responded differently. In this instance, one batch showed some osteoinductive activity, but the other batch showed none.

They came to the conclusion that assays must be developed to standardize DFDBA activity. Studies by Shigeyama et al. and Becker et al. support these findings. Shigeyama et al. evaluated the biological activity of protein extracts generated from freshly acquired human bone and commercially supplied DFDBA in vitro. They discovered that BMP 2, 4, 7, as well as BSP, fibronectin, and type I collagen were present in both commercially available and laboratory-made extracts. Comparing freshly prepared protein extracts to commercially prepared protein extracts, freshly prepared extracts had a higher amount of BMPs and also had a larger capacity to induce cell proliferation. Due to the number of proteins and their biological activity, preparations from commercial laboratories may therefore result in activity loss.

Xenografts

Bones from other animals have long been used in periodontal therapy. Only historical interest is served by mentioning a handful of these xenograft items since they are no longer in use.

Detergent extraction, sterilization, and freeze-drying of calf bone have been employed to cure osseous abnormalities.[81]

Kiel bone is calf or ox bone that has been dried with acetone, sterilized with ethylene oxide, and denatured with 20% hydrogen peroxide.

Anorganic bone is ox bone that has undergone an autoclave process to remove the organic material after utilising ethylenediamine to do so.[82]

Currently, an anorganic, bovine-derived bone has been utilized successfully for implant surgery as well as periodontal problems. It is a porous, osteoconductive bone mineral matrix derived from cortical or cancellous bone in cattle. The trabecular structure and porosity of the bone are preserved, but the organic components are taken out. The physical characteristics enable clot stability and revascularization to enable osteoblast migration and osteogenesis. This transplant doesn't trigger a systemic immune reaction and is biocompatible with the surrounding tissues.[83]

According to Yukna et al., the use of anorganic, bovine-derived bone combined with a cell-binding polypeptide (P-15), a synthetic analogue of a 15-amino acid sequence of type I collagen sold under the name P-15, appears to improve the bone-regenerative effects of the matrix alone in periodontal defects.[84]

Bone Morphogenetic proteins

The bone morphogenetic proteins are a family of proteins found in the body that play an important role in skeletal development. Each of the proteins serves a distinct purpose, and BMP-2 has been shown to have some of the most potent bone-producing activity. Bone formation has also been shown to be stimulated by BMP-7 (also known as osteogenic protein-1, or OP-1) and BMP-3 (also known as osteogenin). Marshall Urist [13] was the first to isolate BMPs from bovine bone. BMPs, as a growth factor, cause mesenchymal stem cells to differentiate into bone-producing osteoblast cells.

Properties of bone morphogenetic proteins

- On undifferentiated mesenchymal cells and osteoblast precursors, they serve as mitogens.
- They are structurally similar TGF-superfamily members.
- BMP 2-12 initiates the development of endochondral bone from scratch.[85-88]
- In contrast to other growth factors like TGF-1 or PDGF, BMPs promote the development of bone.
- BMPs stimulate osteoblastic development in human periodontal ligament (PDL) cells, which has an anabolic effect on periodontal tissue.[89]

- BMPs, such as BMP-2, -4, and -7, are present in different amounts in bone allograft materials.[90] The inability of the bone to heal due to a lack of BMP-like proteins, which slows down bone cell differentiation.[91]
- Studies have demonstrated that recombinant BMPs (rh BMPs) encourage bone growth.[92,93]
- They cause the osteoblast phenotype to be expressed (i.e. increase in alkaline phosphatase activity in bone cells).
- Bind to extracellular matrix collagen type IV and serve as chemoattractants for mesenchymal cells and monocytes.[94]

Structure of BMPs

The BMPs are homodimers of 30- to 38-kDa glycosylated proteins. A cell produces the individual BMP proteins, which dimerize and become glycosylated. They are produced as prepropeptides with 400 to 525 amino acids. The homology of the amino acid sequences was used to divide the BMPs into subsets. The groupings are suggested to be as follows:

(1) BMP-2 and BMP-4,

(2) BMP-3 and BMP-3b,

(3) BMP-5, BMP-6, BMP-7, and BMP-8,

(4) BMP-9 and BMP-10,

(5) BMP-12, BMP-13, and BMP-14, and

(6) BMP-11 and growth/differentiation factor 8 (GDF-8).

Here is a brief explanation of a few thoroughly researched BMPs:

BMP-1 On chromosome 8 is the BMP 1 genes. It is not a member of the family of proteins known as TGF-. Procollagen I, II, and III are affected by this metalloproteinase. It affects how cartilage develops. BMP-2 It is a significant osteoblast differentiation inducer. On chromosome 20, the gene is located. Bone development is induced by BMP-3. The chromosome 14 region contains the genes. BMP-4 controls the mesodermal development of teeth, limbs, and bone. Additionally, it helps to heal

fractures. The fourteenth chromosome contains genes. BMP-5 plays a part in the growth of cartilage. One can find genes on chromosome 6. Adult joint integrity is affected by BMP-6. One can find genes on chromosome 6. BMP-7 is important for osteoblast differentiation.

Role of BMPs in periodontal regeneration

In several animal models,[95-98] as well as in human investigations,[99,100] BMPs have been employed extensively by researchers to stimulate periodontal tissue regeneration with varied degrees of success. According to research, BMPs have a structure/activity profile, with BMP-2 mostly exhibiting osteogenic capabilities and BMP-7 primarily demonstrating cementogenic activities.[101]

Periodontal regeneration has been studied using rhBMP-2, a recombinant human bone morphogenetic protein. Using rhBMP-2 and a synthetic carrier, Sigurdsson et al. (1995)[95] and Kinoshita et al. (1997)[98]" effectively regenerated periodontal tissue in dogs. The protein and the carrier were well tolerated, both locally and systemically, in clinical trials using rhBMP-2 in an absorbable collagen sponge carrier. [99,100]

When injected into extraskeletal locations in several animal models, recombinant human BMP-2 through BMP- 6 and osteogenic protein-1 and -2 (OP-1 and OP-2, also known as BMP-7 and BMP-8, respectively) alone cause de novo bone formation.[102,103] Both a mammalian cell expression system and an Escherichia coli expression system were used to create these recombinant BMPs.

Given that both BMP-2 and BMP-7 expression have been observed during periodontal tissue morphogenesis, it may be necessary to combine the two BMPs for the best possible treatment regeneration. Recent studies have focused on the use of BMPs in regenerative periodontal therapy to promote bone healing. In one study, it was discovered that rhBMP-7 and rhBMP-2 were both secure and efficient in enhancing and speeding up fibrous nonunion fracture healing as well as bone healing in orthotropic animal models.[104] One of the most crucial tasks in the therapeutic application of BMPs is to transfer them through a carrier into a periodontal lesion. The osteoinductive property of rhBMP has been investigated utilising Ca-P-coated porous titanium fibre mesh loaded with rhBMP-2 in subcutaneous implants in rats.[105] Within 7-9 days, ectopic bone creation with a cartilaginous phase was seen, and it was found that the

process resembled endochondral ossification. RhBMP-2 integrated into Ca-P coatings had a stronger capacity to promote alkaline phosphatase activity, which is indicative of bone formation, according to another study on rat bone marrow stromal cells. The surfaces of alveolar bone, cementum, and PDL fibre bundles have been found to contain additional BMP family members, such as growth and differentiation factor-5, 6, and 7. Further research is needed for therapeutic applications.[106]

The current rhBMP-2 technology is produced in a recombinant expression system using Chinese hamster ovarian (CHO) cells that have been genetically modified to over express the BMP-2 coding sequence. This technology is approved for use in the treatment of spine, long-bone fracture, and alveolar augmentation indications. Escherichia coli can also produce rhBMP-2 as inclusion bodies, resulting in high yields and up to 99% purification. In mouse screening models, it has been demonstrated that rhBMP-2 produced from E. coli and CHO cells induce equivalent dose-dependent ectopic bone formation after intramuscular implantation, indicating that rhBMP-2 derived from E. coli is a viable alternative to rhBMP-2 derived from CHO cells. However, there hasn't been much research done on the effectiveness of rhBMP-2 produced from E. coli in large animal, orthotopic, therapeutically relevant settings. So, using an established large animal model, the goal of this work was to assess local bone production, dental implant osseointegration, and alveolar augmentation using rhBMP-2 obtained from E. coli and compares it to rhBMP-2 derived from CHO-cells as a reference.

The first E.rhBMP-2 in the world is a growth factor that promotes the production of bone and cartilage. It is generated from E. coli. The differentiation of osteoblasts is significantly influenced by this retinoid mediator.

The present study is to evaluate both clinical and radiographic bone fill of intrabony defects treated with DFDBA alone and DFDBA in combination with rhBMP-2. This study examined soft tissue changes (probing depth and clinical attachment level) and Plaque index and gingival index in intrabony defects treated with DFDBA alone and DFDBA in combination with rhBMP-2.

This study showed that both treatment modalities resulted in significant improvements in hard and soft tissue measurements. There were significant differences in clinical and radiographic outcomes between the two treatment groups.

DFDBAs have been used for some time in periodontics to try to stimulate bone formation. One mechanism proposed for the osteoinductive ability of DFDBA is the presence of BMPs remaining within the organic matrix after the demineralization process. Interestingly, few studies have analyzed the protein content and composition of commercially available DFDBA preparations; thus, the actual nature of the osteoinductive agents within these preparations is largely unknown. Of even greater interest is the reported variability in osteoinductive capacity of DFDBA preparations. Recently, some of this variation has been attributed to the donor age of the DFDBA specimens.

Plaque index and gingival index

The plaque and gingival index were assessed at baseline and six months to monitor the patient's oral hygiene and its effects on soft tissue, as this helps to achieve the desired goal. The findings of our study revealed a statistically significant decrease in the plaque index from baseline and at the end of six months in both Group A and Group B, which is consistent with a study that concluded that ABBM + GTR (anorganic bovine bone mineral + guided tissue regeneration) with a non-resorbable barrier, with or without the addition of PRP, produced optimal clinical results.[107]

In our study we found that at baseline and 6 months post operative assessment of Plaque Index in Group A showed a significant reduction of 1.5000. Similar result was found for Group B which showed a significant reduction of 1.5000, significant at p = 0.000. Plaque Index after 6 months of intervention reduced to 1.000 + 0.000 in both groups.

Group A had a GI of .5700 $\pm$ .2626 and Group B had .4700 $\pm$ .27101, after 6 months which was not significant. Gingival Index significantly reduced in Group A subjects from 2.5800$\pm$.24404 to .5700 $\pm$ .26268 from baseline to 6 months of intervention. Gingival Index significantly reduced in Group B subjects from 2.500 $\pm$.28284 to .4700 $\pm$ .27101 from baseline to 6 months of intervention.

Individuals' behaviour can be influenced by the feeling of being watched or simply participating in an experiment. This inconsistently observed phenomenon, known as the Hawthorne effect, can both provide insight into individuals' behaviour and confound

the interpretation of experimental manipulations. Therefore the reduction in PI and GI scores can also be attributed to the hawthorne effect[108]

PPD and CAL

The changes in PPD reflect the cumulative effect of the response of gingival tissue to the treatment by way of gingival recession and clinical attachment gain. PPD indicates the volume of subgingival area, which harbours the pathogenic microbiota and favors disease activity. Change in CAL following regenerative therapy is the single most commonly used outcome measure in regenerative therapy. This is based on reported correlation between gain in CAL and gain in bone height by various clinical studies[109].

At the end of six months, reduction in PPD in both groups was at par with each other for Group A and Group B. Periodontal pocket depth exhibited significant reduction from baseline score of 7.000 ± .94231 to 3.4000 ± .69921 after 6 months, significant at p = 0.000 in Group A participants. Periodontal pocket depth exhibited significant reduction from baseline score of 7.000 ± .81650 to 3.4000 ± .69921 after 6 months, significant at p = 0.000 in Group B participants.

When compared between the groups Group A performed slightly better with CAL scoring to 2.600 +.84327 as against 3.3000 + .82327 in Group B, but the difference was not significant. A significant reduction was noted in Clinical attachment level in group A from 6.5000 ± .84984 to 2.6000 ± .84327 at 6 months of intervention which was significant at p=0.000. A significant reduction was noted in Clinical attachment level in group B from 6.7000 ± .82327 to 3.3000 ± .82327 at 6 months of intervention which was significant at p=0.000. The results of our study showed a CAL gain both in group A and B which was statistically significant. This is in accordance with the study where they concluded that both scaling and root planing alone and scaling and root planing combined with flap procedure are effective methods for the treatment of chronic periodontitis in terms of attachment level gain and reduction in gingival inflammation.[110]

It has been reported that rhBMP-2 has the potential to regenerate cement and periodontal ligament[111]. It was discovered that rhBMP-2-regenerated cementoid tissue inhibited epithelial migration. However, Sigurdsson et al. found that rhBMP-2 caused ankylosis in periodontal regeneration sites. It has been reported that a high dose of

rhBMP-2 can cause dentinal resorption. Several studies have found that rhBMP-2 has a bone regeneration effect. In contrast, there is debate about periodontal tissue regeneration.[112]

Bone Gain

When assessed for bone gain between groups, Group A showed bone gain of 2.4490 ± .34719 while it was 4.0880 ± .40105 in the Group B which was statistically significant at p=0.000 A number of studies[17,18]have shown that when added to a variety of carriers and implanted orthotopically, BMP can promote new bone formation. The current study confirms previous findings using guanidine-extracted laboratory-prepared demineralized bone[113], demonstrating that DFDBA is an excellent carrier for BMP, particularly rhBMP-2[114], because new bone formation is induced heterotopically. Indeed, the findings show that when combined with rhBMP-2, even commercial preparations of human DFDBA can effectively promote bone formation.

In this study, rhBMP-2 was more effective as a bone inducing agent than DFDBA alone. Even low doses of rhBMP-2 may have exceeded the amount of BMP present in the active DFDBA preparations, making the composite more inductive. It is impossible to say because we did not measure the absolute BMP content of the DFDBA preparations or investigate the effect of adding rhBMP-2 to the DFDBA. BMP-2 may be stored in bone in an inactive form, with only a portion of it being activated during the acid extraction protocol used to demineralize freeze dried bone. Furthermore, other BMPs and TGFß may be present in bone, which could mitigate the bone induction process. The release of bioactive agents from DFDBA is dependent on macrophage résorption.[115,116] In contrast, all of the rh-BMP-2 mixed with the DFDBA is already inactive; additionally, it is only adsorbed onto the material's surface, resulting in very different release kinetics. Interestingly, rhBMP-2 not only increased bone formation but also appeared to increase the résorption of inactive DFDBA to levels comparable to active DFDBA. This suggests that BMPs may also be involved in the regulation of resorption. It is unknown whether this is accomplished through direct action on resorptive cells, activation of more osteoblasts to produce factors that stimulate osteoclasts, or indirectly through the production of hemapoietic bone marrow. Concerns have also been raised that when DFDBA is used clinically, it is frequently not fully resorbed, potentially altering the release and activity of adsorbed rhBMP-2. Despite

these limitations, our findings suggest that by incorporating rhBMP-2 into commercially available DFDBA, all DFDBA batches could be made active with high predictability. In comparison, the currently available DFDBA has a low level of reliable osteoinductiveability. These findings also suggest that some commercial preparations of DFDBA are inactive in terms of bone induction ability because they lack adequate amounts of BMP.

In recent clinical studies, the adverse effect of rhBMP-2 on facial swelling was reported to be proportional to the dose of rhBMP-2.[117] Because of the low dose of rhBMP-2 used in this study, there were no adverse effects.

The use of bone substitutes in periodontal regeneration therapy is currently a key area of periodontology research. We went into great length about the many types of bone transplants and their sources in the discussion above. Although autogenous bone transplant is the closest to the ideal bone graft, harvesting the graft requires a second surgical site. Although they offer an alternative to autografts, allografts do not have as much of an osteoinductive effect. Composite bone grafts, which combine the qualities of the scaffold and physiologically active molecules such bone morphogenetic proteins/growth factors, are the material of the future for bone grafts. In other words, the graft material serves as a vehicle for molecules that are biologically active. These biomaterials are perfect for transporting these biologically active compounds since they contain collagen matrix. These materials may be the most accurate substitutes for autografts, despite the fact that research into them is still in its early stages.

The clinical application of BMPs in periodontal regeneration has shown promising results, however, a lot of research work is required to fabricate an appropriate carrier system for BMPs. Furthermore, we need to develop cost effective and easily available carrier systems for BMPs so that these can be widely used in different kinds of regenerative procedures.

CONCLUSION

CONCLUSION

The periodontium that has been damaged by periodontal disease may fully recover with periodontal bone grafts. Periodontal therapy's major objective continues to be the regeneration of missing supporting tissues.

On a medium to long term basis, periodontal regenerative procedures, particularly combination techniques comprising DFDBA and rh-BMP-2 grafting, produce clinical and radiographic outcomes in intrabony lesions that are noticeably better than DFDBA. This results in longer-term tooth retention that is why periodontal regenerative/reconstructive therapy is highly advised for the treatment of intrabony abnormalities.

The 2 treatment modalities (Group A & B) showed favorable clinical results. DFDBA with rhBMP-2 showed better results in comparison with DFDBA alone. In our study we observed that the BMPs in DFDBA are somewhat in an inactive form and the addition of rhBMP-2 to DFDBA attained better results.

DFDBA and rhBMP-2 application is of great interest to the researcher as well as clinician, holds promise and needs further exploration. Periodontal maintenance is crucial for any therapy and plays a key role in the long-term prognosis.

More studies with a larger sample size and longer follow up period are required to substantiate the results obtained in this clinical study.

BIBLIOGRAPHY

BIBLIOGRAPHY

1. Bosshardt DD, Sculean A (2009) Does periodontal tissue regeneration really work? Periodontol 2000 51: 208-19.
2. Khajuria DK, Zahra SF, Razdan R (2018) Effect of locally administered novel biodegradable chitosan based risedronate/zinc-hydroxyapatite intra-pocket dental film on alveolar bone density in rat model of periodontitis. J Biomater Sci Polym Ed 29: 74-91.
3. Melcher AH (1976) On the repair potential of periodontal tissues. J Periodontol 47: 256-60.
4. Caton JG, Greenstein G (1993) Factors related to periodontal regeneration. Periodontol 2000 1: 9-15.
5. Bartold PM (2015) Group C. Initiator paper. Periodontal regeneration–fact or fiction. J Int Acad Periodontol 17: 37-49.
6. Bröseler F, Tietmann C, Hinz AK, Jepsen S, Čebatariūnienė A, et al. (2005) Position paper: Periodontal regeneration. J Periodontol 76: 1601-22.
7. Gottlow, J., Nyman, S., Lindhe, J., Karring, T., Wennström, J. New attachment formation in the human periodontium by guided tissue regeneration. Case reports. J Clin Periodontol. 1986; 13: 604-16.
8. Hiatt, W. H., Schallhorn, R. G., Aaronian, A. J. The induction of new bone and cementum formation. IV. Microscopic examination of the periodontium following human bone and marrow allograft, autograft and nongraft periodontal regenerative procedures. J Periodontol. 1978; 49: 495-512.
9. Tonetti, M. S., Pini, P. G., Williams, R. C., Cortellini, P. Periodontal regeneration of human infrabony defects. III. Diagnostic strategies to detect bone gain. J Periodontol. 1993; 64: 269-77.
10. Bowers, G. M., Chadroff, B., Carnevale, R., Mellonig, J., Corio, R., Emerson, J., Stevens, M., Romberg, E. Histologic evaluation of new attachment apparatus formation in humans. Part III. JPeriodontol. 1989; 60: 683-93.
11. Mellonig, J. T., Bowers, G. M., Cotton, W. R. Comparison of bone graft materials. Part II. New bone formation with autografts and allografts: a histological evaluation. J Periodontol. 1981; 52: 297-302.
12. Piorellini, M., Nevins, A. Localized Ridge Augmentation/Preservation. A systemic review. Periodontol. 2003; 8: 321-327.

13. Urist MR (1965) Bone: formation by autoinduction. *Science* 150: 893-899

14. Urist MR, Strates BS (1971) Bone morphogenetic protein.] *Dent Res 50:1392-1406*

15. Shigeyama Y, D'Errico JA, Stone R, Somerman MJ (1995) Commercially-prepared allograft material has biological activity *in vitro.*] *Periodontol66: 478-487.*

16. Schwartz Z, Mellonig JT, Carnes D L, Delafontaine J, Cochran D L, Dean DD, Boyan BD (1996) Ability of commercial demineralized freeze-dried bone allograft to induce new bone formation.] *Periodontol67: 918-926*

17. Becker W, Urist MR, Tucker LM, Becker BE, Ochsenbein C (1995) Human demineralized freeze-dried bone: inadequate induced bone formation in athymic mice. A preliminary report.] *Periodontol* 66: 822-828.

18. Urist, M. R. Bone: formation by autoinduction. Science. 1965; 150: 893–899.

19. Wozney, J. M., Rosen, V., Celeste, A. J., Mitsock, L. M., Whitters, M. J., Kriz, R. W., Hewick, R. M., Wang, E. A. Novel regulators of bone formation: molecular clones and activities. Science. 1988; 242: 1528–1534.

20. Celeste, A. J., Iannazzi, J. A., Taylor, R. C., Hewick, R. M., Rosen, V., Wang, E. A. and Wozney, J. M. Identification of transforming growth factor b family members present in boneinductive protein purified from bovine bone. Proceedings of the National Academy of Sciences of the United States of America. 1990; 87: 9843–9847.

21. Özkaynak, E., Rueger, D. C., Drier, E. A., Corbett, C., Ridge, R. J., Sampath, T. K. and Oppermann, H. OP-1 cDNA encodes an osteogenic protein in the TGF-b family. EMBO Journal. 1990; 9: 2085–2093.

22. Wang, E. A., Rosen, V., D'Alessandro, J. S., Bauduy, M., Cordes, P., Harada, T., Israel, D. I., Hewick, R. M., Kerns, K. M., LaPan, P., Luxenburg, D. P., McQuaid, D., Moutsatsos, I. K., Nove, J., and Wozney, J. M. Recombinant human bone morphogenetic protein induces bone formation. Proceedings of the National Academy of Sciences of the United States of America. 1990; 87: 2220–2224.

23. Katagiri, T., Takahashi, N. Regulatory mechanisms of osteoblast and osteoclast differentiation. Oral Dis. 2002; 8: 147–159.

24. Canalis, E., Economides, A. N., Gazzerro, E. Bone morphogenetic proteins, their antagonists, and the skeleton. Endocr. Rev. 2003; 24: 218–235.

25. Wikesjö, U. M., Lim, W. H., Thomson, R. C., Cook, A. D., Wozney, J. M., Hardwick, W. R. Periodontal repair in dogs: evaluation of a bioresorbable space-providing macroporous membrane with recombinant human bone morphogenetic protein-2. Journal of Periodontology. 2003; 74: 635–647.

26. Wikesjö, U. M., Xiropaidis, A. V., Thomson, R. C., Cook, A. D., Selvig, K. A. and Hardwick, W. R. Periodontal repair in dogs: rhBMP-2 significantly enhances bone formation under provisions for guided tissue regeneration. Journal of Clinical Periodontology. 2003b; 30: 705–714.

27. Yudell, R. M., Block, M. S. Bone gap healing in the dog using recombinant human bone morphogenetic protein-2. Journal of Oral and Maxillofacial Surgery. 2000; 58: 761–766.

28. Hunt, D. R., Jovanovic, S. A., Wikesjö, U. M., Wozney, J. M. and Bernard, G. W. Hyaluronan supports recombinant human bone morphogenetic protein-2 induced bone reconstruction of advanced alveolar ridge defects in dogs. A pilot study. Journal of Periodontology. 2001; 72: 651–658.

29. Sykaras, N., Triplett, R. G., Nunn, M. E., Iacopino, A. M. and Opperman, L. A. Effect of recombinant human bone morphogenetic protein-2 on bone regeneration and osseointegration of dental implants. Clinical Oral Implants Research. 2001; 12: 339–349.

30. Jovanovic, S. A., Hunt, D. R., Bernard, G. W., Spiekermann, H., Wozney, J. M. and Wikesjö, U. M. Long-term functional loading of dental implants in rhBMP-2 induced bone. A histologic study in the canine ridge augmentation model. Clinical Oral Implants Research. 2003; 14: 793–803.

31. Nakashima, K., Zhou, X., Kunkel, G., Zhang, Z., Deng, J. M., Behringer,R. R. and Crombrugghe, B. The novel zinc finger-containing transcription factor osterix is required for osteoblast differentiation and bone formation. Cell. 2002; 108: 17–29.

32. Mellonig JT, Bowers GM, Cotton WR. Comparison of bone graft materials: Part II. New bone formation with autografts and allografts: a histological evaluation. Journal of Periodontology. 1981 Jun;52(6):297-302.

33. Toriumi DM, Kotler HS, Luxenberg DP, Holtrop ME, Wang EA. Mandibular reconstruction with a recombinant bone-inducing factor: Functional, histologic, and biomechanical evaluation. Archives of Otolaryngology–Head & Neck Surgery. 1991 Oct 1;117(10):1101-12.

34. Anderegg CR, Martin SJ, Gray JL, Mellonig JT, Gher ME. Clinical evaluation of the use of decalcified freeze-dried bone allograft with guided tissue regeneration in the treatment of molar furcation invasions. Journal of periodontology. 1991 Apr;62(4):264-8

35. Garraway R, Young WG, Daley T, Harbrow D, Bartold PM. An assessment of the osteoinductive potential of commercial demineralized freeze-dried bone in the murine thigh muscle implantation model. Journal of periodontology. 1998 Dec;69(12):1325-36.

36. Schwartz Z, Somers A, Mellonig JT, Carnes Jr DL, Wozney JM, Dean DD, Cochran DL, Boyan BD. Addition of human recombinant bone morphogenetic protein-2 to inactive commercial human demineralized freeze-dried bone allograft makes an effective composite bone inductive implant material. Journal of periodontology. 1998 Dec;69(12):1337-45.

37. Li H, Pujic Z, Xiao Y, Artold PM. Identification of bone morphogenetic proteins 2 and 4 in commercial demineralized freeze-dried bone allograft preparations: pilot study. Clinical Implant Dentistry and Related Research. 2000 Apr;2(2):110-7.

38. Jepsen S, Terheyden H. Bone morphogenetic proteins in periodontal regeneration. InBone morphogenetic proteins 2002 (pp. 183-192). Birkhäuser, Basel.

39. Jovanovic SA, Hunt DR, Bernard GW, Spiekermann H, Nishimura R, Wozney JM, Wikesjö UM. Long-term functional loading of dental implants in rhBMP-2 induced bone: A histologic study in the canine ridge augmentation model. Clinical oral implants research. 2003 Dec;14(6):793-803

40. Jung RE, Glauser R, Schärer P, Hämmerle CH, Sailer HF, Weber FE. Effect of rhBMP-2 on guided bone regeneration in humans: A randomized, controlled clinical and histomorphometric study. Clinical oral implants research. 2003 Oct;14(5):556-68.

41. Xiao YT, Xiang LX, Shao JZ. Bone morphogenetic protein. Biochemical and biophysical research communications. 2007 Oct 26;362(3):550-3.

42. Lan J, Wang ZF, Shi B, Xia HB, Cheng XR. The influence of recombinant human BMP-2 on bone–implant osseointegration: biomechanical testing and histomorphometric analysis. International journal of oral and maxillofacial surgery. 2007 Apr 1;36(4):345-9.

43. Wikesjö UM, Huang YH, Polimeni G, Qahash M. Bone morphogenetic proteins: a realistic alternative to bone grafting for alveolar reconstruction. Oral and maxillofacial surgery clinics of North America. 2007 Nov 1;19(4):535-51

44. Piemontese M, Aspriello SD, Rubini C, Ferrante L, Procaccini M. Treatment of periodontal intrabony defects with demineralized freeze-dried bone allograft in combination with platelet-rich plasma: A comparative clinical trial. Journal of periodontology. 2008 May;79(5):802-10.

45. King GN, King N, Hughes FJ. Effect of two delivery systems for recombinant human bone morphogenetic protein-2 on periodontal regeneration in vivo. Journal of periodontal research. 1998 Apr;33(3):226-36.

46. Thoma DS, Jones A, Yamashita M, Edmunds R, Nevins M, Cochran DL. Ridge augmentation using recombinant bone morphogenetic protein-2 techniques: an experimental study in the canine. Journal of periodontology. 2010 Dec;81(12):1829-38

47. Bashutski JD, Wang HL. Biologic agents to promote periodontal regeneration and bone augmentation. Clinical Advances in Periodontics. 2011 Aug;1(2):80-7.

48. Spagnoli DB, Marx RE. Dental implants and the use of rhBMP-2. Dental Clinics. 2011 Oct 1;55(4):883-907.

49. Khojasteh A, Behnia H, Naghdi N, Esmaeelinejad M, Alikhassy Z, Stevens M. Effects of different growth factors and carriers on bone regeneration: a systematic review. Oral surgery, oral medicine, oral pathology and oral radiology. 2013 Dec 1;116(6):e405-23.

50. SINGH GR. BoneMorphogenic Protein-Ashort communication. CODS-Journal of Dentistry. 2013 Sep 1;2(1):1-3.

51. Hur BM, Lim SB. The effect of rhBMP-2 bonegraft on infrabony defects

52. Chadwick JK, Mills MP, Mealey BL. Clinical and radiographic evaluation of demineralized freeze-dried bone allograft versus platelet-rich fibrin for the treatment of periodontal Intrabony defects in humans. Journal of periodontology. 2016 Nov;87(11):1253-60.

53. Jaiswal Y, Kumar S, Mishra V, Bansal P, Anand KR, Singh S. Efficacy of decalcified freeze-dried bone allograft in the regeneration of small osseous defect: A comparative study. National Journal of Maxillofacial Surgery. 2017 Jul;8(2):143.

54. Schorn L, Sproll C, Ommerborn M, Naujoks C, Kübler NR, Depprich R. Vertical bone regeneration using rhBMP-2 and VEGF. Head & Face Medicine. 2017 Dec;13(1):1-1.

55. Bavsar AK, Prabhuji ML, Varadhan KB, Parween S. Critical issues in periodontal regeneration-A Review. J Oral Health Dent. 2018;2:204

56. Petsos H, Ratka-Krüger P, Neukranz E, Raetzke P, Eickholz P, Nickles K. Infrabony defects 20 years after open flap debridement and guided tissue regeneration. Journal of Clinical Periodontology. 2019 May;46(5):552-63.

57. Alhussaini AH. Effect of platelet-rich fibrin and bone morphogenetic protein on dental implant stability. Journal of Craniofacial Surgery. 2019 Jul 1;30(5):1492-6.

58. Atchuta A, Gooty JR, Guntakandla VR, Palakuru SK, Durvasula S, Palaparthy R. Clinical and radiographic evaluation of platelet-rich fibrin as an adjunct to bone

grafting demineralized freeze-dried bone allograft in intrabony defects. Journal of Indian Society of Periodontology. 2020 Jan;24(1):60.

59. Löe H. The Gingival Index, the Plaque Index and the Retention Index Systems. J Periodontol. 1967 Nov-Dec;38(6):Suppl:610-6.

60. Hefti AF. Periodontal probing. Crit Rev Oral Biol Med. 1997;8(3):336-56.

61. Pihlstrom BL. Measurement of attachment level in clinical trials: probing methods. Journal of periodontology. 1992 Dec;63:1072-7.

62. Ellegaard, B. & Loe, H. (1971). New attachment of periodontal tissues after treatment of intrabony lesions. *Journal of Periodontology* **42**, 648–652.

63. Ellegaard, B., Karring, T., Davies, R. & Loe, H. (1974). New attachment after treatment of intrabony defects in monkeys. *Journal of Periodontology* **45**, 368–377.

64. Ellegaard, B., Karring, T. & Loe, H. (1975). The fate of vital and devitalized bone grafts on the healing of interradicular lesion. *Journal of Periodontal Research* **10**, 88–97.

65. Ellegaard, B., Nielsen, I.M. & Karring, T. (1976). Composite jaw and iliac cancellous bone grafts in intrabony defects in monkeys. *Journal of Periodontal Research* **11**, 299–310.

66. Abbott LC, Schottstaedt ER, SAUNDERS JB, Bost FC. The evaluation of cortical and cancellous bone as grafting material: A clinical and experimental study. JBJS. 1947 Apr 1;29(2):381-414.

67. Wilk RM. Bony reconstruction of the jaws. In: Miloro M, editor. Peterson's principles of oral and maxillofacial surgery. 2nd edition. Hamilton (London): B C Decker; 2004. Pp. 785–7.

68. Hegedus Z. The rebuilding of the alveolar processes by bone transplantation. Den. Cosmos. 1923;65:736-42.

69. O'Leary TJ, Rudd K. D., and Nabers, CL: Factors affecting horizontal tooth mobility. SAM-TR-65-19, April; 1965

70. Nabers CL. Long-term results of autogenous bone grafts. Int Periodont Restorative Dent 1984 4 (3): 50-57.

71. Froum SJ, Thaler R, Scoop IW, et al: Osseous autografts. I. Clinical responses to bone blend or hip marrow grafts. J Periodontol 46:515, 1975.

72. Ewen SJ: Bone swaging. J Periodontol 36:57, 1965

73. Russo R, Scarborough N. Inactivation of viruses in demineralized bone matrix. FDA Workshop on Tissue for Transplantation and Reproductive Tissue. June 20-21, 1995, Bethesda, MD.

74. Cushing M. Autogenous red marrow grafts: their potential for induction of osteogenesis. J Periodontol 1969;40(8):492-7.

75. Sottosanti JS, Bierly JA. The storage of bone marrow and its relation to periodontal grafting procedures. J Periodontol 1975:46 (3):162-70.

76. Amler MH. The effectiveness of regenerating versus mature marrow in physiologic autogenous transplants. J Periodontol 1984; 55(5):268-72.

77. Rosen PS, Reynolds MA, Bowers GM. The treatment of intrabony defects with bone grafts. Periodontol 2000 2000;22(1):88-103.

78. Centers for Disease Control and Prevention. Bone allografts. What is the risk of disease transmission with bone allografts? In: Department of Health and Human Services. Available at: http://www.ed.gov/OralHealth/Infectioncontrol/faq/allografts.htm Accessed 19 Oct 2010.

79. Joyce MJ. Safety and FDA regulations for musculoskeletal allografts: perspective of an orthopaedic surgeon. Clin Orthop Relat Res 2005;435:22-30.

80. Vangsness CT, Garcia IA, Mills CR, Kainer MA, Roberts MR. Moore TM. Allograft transplantation in the knee: tissue regulation, procurement, processing, and sterilization. Am J Sports Med 2003; 31(3):474-81.

81. Schaffer EM: Cartilage grafts in human periodontal pockets. J Periodontol 29:176, 1958.

82. Mellado JR, Salkin LM, Freedman AL, et al: A comparative study of ePTFE membranes with and without decalcified freeze-dried bone allografts for the regeneration of interproximal intraosseous defects. J Periodontol 66:751, 1995.

83. Melcher AH: On repair potential of periodontal tissues. J Periodontol 47:256, 1976.

84. Yukna RA, Krauser JT, Callan DP, et al: Multi-center clinical comparison of combination anorganic bovine-derived hydroxyapatite matrix (ABM)/cell binding peptide (P-15) and ABM in human periodontal osseous defects: 6-month results. J Periodontol 71:1671, 2000.

85. Urist MR. Bone: formation by autoinduction. Science 1965,150(3698):893-9.

86. Celeste AJ, lannazzi JA, Taylor RC, Hewick RM, Rosen V, Wang EA, Wozney JM. Identification of transforming growth factor beta family members present in bone-

inductive protein purified from bovine bone. Proc Natl Acad Sci USA 1990;87(24):9843-7.

87. Ozkaynak E, Rueger DC, Drier EA, Corbett C, Ridge RJ, Sampath TK, Oppermann H. OP-1 cDNA encodes an osteogenic protein in the TGF-beta family. EMBO J 1990;9(7):2085.

88. Ozkaynak E, Schnegelsberg PN, Jin DF, Clifford GM, Warren FD, Drier EA, Oppermann H. Osteogenic protein-2. A new member ofthe transforming growth factor-beta superfamily expressed early in embryogenesis. Biol Chem 1992;267(35):25220-7.

89. Eickholz P, Krigar DM, Kim TS, Reitmeir P, Rawlinson A. Stability of clinical and radiographic results after guided tissue regeneration in infrabony defects. J Periodontol 2007;78(1):37-46.

90. Shigeyama Y, D'Errico JA, Stone R, Somerman MJ. Commercially- prepared allograft material has biological activity in vitro. J Periodontol 1995;66(6):478-87.

91. Urist MR, DeLange RJ, Finerman GA. Bone cell differentiation and growth factors. Science 1983;220(4598):680-6.

92. Yasko AW, Lane JM, Fellinger EJ, Rosen V, Wozney JM, Wang EA. The healing of segmental bone defects, induced by recombinant human bone morphogenetic protein (rhBMP-2). A radiographic, histological, and biomechanical study in rats. J Bone Joint Surg Am 1992;74(5):659-70

93. Ripamonti U, Ma SS, Cunningham NS, Yeates L, Reddi AH. Reconstruction of the bone-bone marrow organ by osteogenin, a bone morphogenetic protein, and demineralized bone matrix in calvarial defects of adult primates. Plast Reconstr Surg 1993; 91(1):27-36.

94. Paralkar VM, Nandedkar AK, Pointer RH, Kleinman HK, Reddi AH. Interaction of osteogenin, a heparin binding bone morpho- genetic protein, with type IV collagen. J Biol Chem 1990; 265(28): 17281-4.

95. Sigurdsson TJ, Lee MB, Kubota K, Turek TJ, Wozney JM, Wikesjö UM. Periodontal repair in dogs: recombinant human bone morpho- genetic protein-2 significantly enhances periodontal regeneration. J Periodontol 1995;66(2):131-8.

96. Nevins M, Kirker-Head C, Nevins M, Wozney JA, Palmer R, Graham D. Bone formation in the goat maxillary sinus induced by absorbable collagen sponge implants impregnated with recombi- nant human bone morphogenetic protein-2. Int J Periodontics Restorative Dent 1996;16(1):8-19.

97. Ripamonti U, Reddi AH. Tissue engineering, morphogenesis, and regeneration of the periodontal tissues by bone morphogenetic proteins. Crit Rev Oral Biol Med 1997;8(2):154-63.

98. Kinoshita A, Oda S. Takahashi K, Yokota S, Ishikawa I. Periodontal regeneration by application of recombinant human bone morphogenetic protein-2 to horizontal circumferential defects created by experimental periodontitis in beagle dogs. J Periodontol 1997;68(2):103-9.

99. Howell TH, Fiorellini J, Jones A, Alder M, Nummikoski P, Lazaro M. Lilly L, Cochran D. A feasibility study evaluating rhBMP- 2/absorbable collagen sponge device for local alveolar ridge preservation or augmentation. Int J Periodontics Restorative Dent 1997:17(2):124-39.

100. Cochran DL, Jones AA, Lilly LC, Fiorellini JP, Howell H. Evaluation of recombinant human bone morphogenetic protein-2 in oral applications including the use of endosseous implants: 3-year results of a pilot study in humans. J Periodontol 2000;71(8):1241- 57.

101. Ripamonti U, Teare J, Petit JC. Pleiotropism of bone morphogenetic proteins: from bone induction to cementogenesis and periodontal ligament regeneration. J Int Acad Periodontol 2006;8(1):23-32.

102. Reddi AH. Regulation of cartilage and bone differentiation by bone morphogenetic proteins. Curr Opin Cell Biol 1992;4(5):850-5.

103. Reddi AH. Bone and cartilage differentiation. Curr Opin Gen Dev 1994;4(5):737-44.

104. Cook SD, Barrack RL. The use of the OP-1 implant in reconstructive surgery of the hip and knee. In: Fifteen years of clinical experience with hydroxyapatite coatings in joint arthro- plasty 2004; pp. 349-55. Springer Paris.

105. Vehof JW, Mahmood J, Takita H, Hof MV, Kuboki Y, Spauwen PH, Jansen JA. Ectopic bone formation in titanium mesh loaded with bone morphogenetic protein and coated with calcium phosphate. Plast Reconstr Surg 2001;108(2):434-43.

106. Li P. Biomimetic nano-apatite coating capable of promoting bone ingrowth. J Biomed Mater Res A 2003;66(1):79-85. 39. Sena K, Morotome Y, Baba O, Terashima T, Takano Y, Ishikawa I. Gene expression of growth differentiation factors in the developing periodontium of rat molars. J Dent Res 2003;82(3):166-71.

107. Döri F, Huszár T, Nikolidakis D, Arweiler NB, Gera I, Sculean A. Effect of platelet-rich plasma on the healing of intra-bony defects treated with a natural bone mineral and a collagen membrane. J Clin Periodontol 2007; 34(3): 254-61.

108. Schwartz D, Fischhoff B, Krishnamurti T, Sowell F. The Hawthorne effect and energy awareness. Proceedings of the National Academy of Sciences. 2013 Sep 17;110(38):15242-6.

109. Yassibag-Berkman Z, Tuncer O, Subasioglu T, Kantarci A. Combined use of platelet-rich plasma and bone grafting with or without guided tissue regeneration in the treatment of anterior interproximal defects. J Periodontol 2007; 78(5): 801-9.

110. Heitz-Mayfield LJ, Trombelli L, Heitz F, Needleman I, Moles D. A systematic review of the effect of surgical debridement *vs* non-surgical debridement for the treatment of chronic periodontitis. J Clin Periodontol 2002; 29(Suppl. 3): 92-102.

111. Oda, S., Kinoshita, A., Higuchi, T., Shizuya, T., Ishikawa, I. Ectopic bone formation by biphasic calcium phosphate (BCP) combined with recombinant human bone morphogenetic protein-2 (rhBMP-2). J Med Dent Sci. 1997; 44: 53-62.

112. Sigurdsson, T. J., Lee, M. B., Kubota, K., Turek, T. J., Wozney, J. M., Wikesjö, U. M. Periodontal repair in dogs: recombinant human bone morphogenetic protein-2 significantly enhances periodontal regeneration. J Periodontol. 1995; 66: 131-8.

113. Reddi AH, Cunningham NS. Bone induction by osteogenin andbone morphogenetic proteins. [Review]. Biomaterials 1990;11:33—34.

114. Wang EA, Rosen V, D'Alessandro JS, et al. Recombinant humanbone morphogenetic protein induces bone formation. Proc Nati Acad Sci (USA) 1990;87:2220-2224.

115. Reddi AH. Regulation of cartilage and bone differentiation by bone morphogenetic proteins. [Review]. Curr Opin Cell Biol 1992;4:850-855.40.

116. Athanasou NA. Cellular biology of bone-resorbing cells. J Bone Joint Surg 1996;78A: 1096-1112.

117. Boakye, M., Mummaneni, P. V., Garrett M., Rodts, G., and Haid, R. Anterior cervical discectomy and fusion involving a polyetheretherketone spacer and bone morphogenetic protein. J Neurosurg Spine. 2005; 2: 521–525.

ANNEXURE

ANNEXURES

ANNEXURE - 1

Institutional Ethical Committee

Babu Banarasi Das University
Babu Banarasi Das College of Dental Sciences,
BBD City, Faizabad Road, Lucknow – 226028 (INDIA)

Dr. Lakshmi Bala
Professor and Head Biochemistry and
Member-Secretary, Institutional Ethics Committee

Communication of the Decision of the IXth Institutional Ethics Sub-Committee

IEC Code: 09 **BBDCODS/04/2022**

Title of the Project: Comparative Evaluation of the Effectiveness of Demineralized Freeze-Dried Bone Allograft (DFDBA) alone and Demineralized Freeze-Dried Bone Allograft (DFDBA) with Recombinant Human Bone Morphogenetic Protein-2 (rhBMP-2) in the Treatment of Intrabony Defects: A Clinico - Radiographic Study.

Principal Investigator: Dr Ankit Bhadani **Department:** Periodontology

Name and Address of the Institution: BBD College of Dental Sciences Lucknow.

Type of Submission: Revised, MDS Project Protocol

Dear Dr Ankit Bhadani,

The Institutional Ethics Sub-Committee meeting comprising following four members was held on 07th April, 2022.

1.	Dr. Lakshmi Bala Member Secretary	Prof. and Head, Department of Biochemistry, BBDCODS, Lucknow
2.	Dr. Amrit Tandan Member	Prof. & Head, Department of Prosthodontics and Crown & Bridge, BBDCODS, Lucknow
3.	Dr. Rana Pratap Maurya Member	Reader, Department of Orthodontics, BBDCODS, Lucknow
4.	Dr. Akanksha Bhatt Member	Reader, Department of Conservative Dentistry & Endodontics, BBDCODS, Lucknow

The committee reviewed and discussed your submitted documents of the current MDS Project Protocol in the meeting.
The comments were communicated to PI thereafter it was revised.

Decisions: The committee approved the above protocol from ethics point of view.

Forwarded by:

Lakshmi Bala

(Dr. Lakshmi Bala)
Member-Secretary
IEC

Member-Secretary
Institutional Ethic Committee
BBD College of Dental Sciences
BBD University
Faizabad Road, Lucknow-226028

(Dr. Puneet Ahuja)
Principal
BBDCODS

PRINCIPAL
Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)
BBD City, Faizabad Road, Lucknow-226028

ANNEXURE – 2

Institutional research committee approval certificate

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES
(FACULTY OF BBD UNIVERSITY), LUCKNOW

INSTITUTIONAL RESEARCH COMMITTEE APPROVAL

The project titled **"Comparative Evaluation of the Effectiveness of Demineralized Freeze-Dried Bone Allograft (DFDBA) alone and Demineralized Freeze-Dried Bone Allograft (DFDBA) with Recombinant Human Bone Morphogenetic Protein-2 (rhBMP-2) in the Treatment of Intrabony Defects: A Clinico - Radiographic Study"** submitted by **Dr Ankit Bhadani** Post graduate student from the **Department of Periodontology** as part of MDS Curriculum for the academic year 2020-2023 with the accompanying proforma was reviewed by the Institutional Research Committee present on **11th October 2021** at BBDCODS.

The Committee has granted approval on the scientific content of the project. The proposal may now be reviewed by the Institutional Ethics Committee for granting ethical approval.

Prof. Vandana A Pant
Co-Chairperson

Prof. B. Rajkumar
Chairperson

ANNEXURE -3

Consent Form

Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)

BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

Consent Form (English)

Title of the Study: Comparative evaluation of the effectiveness of demineralized freeze- dried bone allograft(DFDBA) alone and demineralized freeze-dried bone allograft(DFDBA) with recombinant human bone morphogenetic protein-2(rhBMP-2) in the treatment of intrabony defects : a clinico - radiographic study

Study Number……..

Subject's Full Name…….

Date of Birth/Age ……

Address of the Subject…….

Phone no. and e-mail address……

Qualification ……

Occupation: Student / Self Employed / Service / Housewife/ Other (Please tick as appropriate)

Annual income of the Subject……

Name and of the nominees(s) and his relation to the subject. (For the purpose of compensation in case of trial related death).

1. I confirm that I have read and understood the Participant Information Document dated ……..for the above study and have had the opportunity to ask questions. **OR** I have been explained the nature of the study by the Investigator and had the opportunity to ask questions.
2. I understand that my participation in the study is voluntary and given with free will without any duress and that I am free to withdraw at any time, without giving any reason and without my medical care or legal rights being affected.
3. I understand that the sponsor of the project, others working on the Sponsor's behalf, the Ethics Committee and the regulatory authorities will not need my permission to look at my health records both in respect of the current study and any further research that may be conducted in relation to it, even if I withdraw from the trial. However, I understand that my Identity will not be revealed in any information released to third parties or published.
4. I agree not to restrict the use of any data or results that arise from this study provided sucha use is only for scientific purpose(s).
5. I permit the use of stored sample (tooth/tissue/blood) for future research. Yes [ ] No [ ] Not Applicable [ ]
6. I agree to participate in the above study. I have been explained about the complications andside effects, if any, and have fully understood them. I have also read and understood the participant/volunteer's Information document given to me.

Signature (or Thumb impression) of the Subject/Legally Acceptable Representative:……..

Signatory‘s Name……. Date …….

Signature of the Investigator…… Date……..

Study Investigator‘s Name..... Date……..

Signature of the witness…… Date……..

Name of the witness……

Received a signed copy of the PID and duly filled consent form

Signature/thumb impression of the subject or legally Date……..

ANNEXURE - 4

PID Form

Babu Banarasi Das College of Dental Sciences

(Babu Banarasi Das University)

BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

Guidelines for Devising a Participant / Legally Acceptable Representative Information Document (PID) in English

1. **Study Title**

   Comparative evaluation of the effectiveness of demineralized freeze- dried bone allograft(dfdba) alone and demineralized freeze-dried bone allograft(DFDBA) with recombinant human bone morphogenetic protein-2(rhBMP-2) in the treatment of intrabony defects : a clinico - radiographic study.

2. **Invitation Paragraph**

   You are being invited to take part in a research study. Before you decide it is important for you to understand why the research is being done and what it will involve. Please take time to read the following information carefully and discuss it with friends, relatives and your treating physician/family doctor if you wish. Ask us if there is anything that is not clear or if you would like more information**.**

3. **What is the purpose of the study?**

   To evaluate and compare the clinical and radiographic outcomes observed in treating intrabony defects with DFDBA alone and DFDBA in conjunction with rhBMP-2

4. **Why have I been chosen?**

   You are chosen as you fulfill the criteria for the study**.**

5. **Do I have to take part?**

   It is up to you to decide whether or not to take part. If you do decide to take part, you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part, you are still free to withdraw at any time and without giving a reason.

6. What will happen to me if I take part?

You will have to come four to five times, in the first visit the tooth will be prepared followed by the measurement of bony defect and placement of bone graft and followed by measurement of defect after 6 months. As a volunteer, your responsibility will be to arrive on time.

7. What do I have to do?

There will be certain changes made in the dietary intake with few other precautionary measures, and you will be expected to follow that.

8. What is the procedure that is being tested?

The defect sites in Group A will be grafted with demineralized freeze- dried bone allograft (DFDBA). The flap will be sutured in close approximation using interrupted sutures. Surgical site will be protected by applying a periodontal dressing.

Similar surgical procedure will be done for Group B. The sites will be grafted with Demineralized freeze-dried bone allograft (DFDBA) in combination with Recombinant human bone morphogenetic protein -2(rhBMP-2).

Further recalls for clinical and radiographic re- evaluation will be schedule at 3 months and 6 months. At each visit, plaque control measures will be reinforced and supra gingival scaling will be done if required.

9. What are the interventions for the study?

Pre-surgical: CBCT will be obtained before starting the procedure Surgical: implant site will be prepared under 2% lignocaine with adrenaline and full thickness of flap will be raised. Then bone graft will be placed, in the defect and we will measure the vertical defect before and after the placement of bone graft and will also be done after 3 month and 6 months . Post-surgical: medications will be prescribed such as: Antibiotics, NSAIDS**.**

10. What are the side effects of taking part?

There are some associated side effects bone graft placement such as pain and discomfort last not more than two weeks, in case of any major problem please report immediately to the

doctor.

11. What are the possible disadvantages and risks of taking part?

- Patients with any systemic diseases that affects the periodontal treatment outcome.
- Pregnant and lactating women.
- Smokers and tobacco chewers.
- Patients who have used antibiotics for the previous 3 months.
- Subjects with a known allergy to the material being used.

12. What are the possible benefits of taking part?

By taking part in this study you will be receiving a better treatment option at a lesser discomfort. These types of bone graft will produce good results since they have growth factors in them.

13. What if new information becomes available?

Sometimes during the course of a research project, new information becomes available about the research being studied. If this happens, you will be informed about it and the changes that can happen to the study will be informed. You are free to withdraw in the middle of the study. If you decide to continue in the study, you may be asked to sign an updated consent form.

14. What happens when the research study stops?

If the study finishes/stops before the stipulated time, then the reason for the same will be explained to the patients.

15. What if something goes wrong?

Volunteers will be taken care of by the doctors expertising in the field at BBDCODS opd.

16. Will my taking part in this study be kept confidential?

Your name, address or any personal or other information will not be shared outside the BBDCODS.

17. What will happen to the results of the research study?

Identity of the participants will not be disclosed in any result/ reports/ publications**.**

18. Who is organizing the research?

Study is organized by the researcher. Complete cost of the bone graft will be given by the patient**.**

19. Will the results of the study be made available after study is over?

If the patient wishes, the result of the study will be made available to him/ her.

20. Who has reviewed the study?

The HOD /IRC/IEC of the institution has reviewed and approved the study

21. Contact for further information

Dr Ankit Bhadani
Department of Periodontology
Address: Banarasi das University, Faizabad road, Atif Vihar, Lucknow, UP. 226028
Email: ankitbhadani.11@gmail.com

Dr. Lakshmi Bala
Member Secretary of Ethics Committee of the institution,
Address: Babu Banarasi das University, Faizabad road, Atif Vihar, Lucknow, UP. 226028 Email: bbdcods.iec@gmail.com

Dr. Suraj Pandey (Reader)
Department of Periodontology and Implantology
Babu Banarasi College of Dental Sciences.
Lucknow-227105
Mob- 9628931689

Dr. Mona Sharma (HOD)
Department of Periodontology and Implantology
Babu Banarasi College of Dental Sciences.
Lucknow-227105
Mob-9984110444

Name of pt. –

Address –

Email –

Tel no. –

Signature of PI……

Name……..

Date……..

The participant will be given a copy of the information sheet and the signed consent form.

Thank you for taking part in the study.

ANNEXURE - 5

बाबूबनारसीदासकॉलेजऑफडेंटलसाइंसेज

(बाबूबनारसीदासविश्वविद्यालयकाएकघटकसंस्थान)

बीबीडीवसटी, फैजाबादरोड, लखनऊ- 227105 (भारत)

प्रतिभागीसूचनादस्तावेज (पीआईडी)

1. **अध्ययन शीर्षक**
   इंट्राबोनी दोषों के उपचार में डिमिनरलाइज्ड फ्रीज-ड्राय बोन एलोग्राफ्ट (डीएफडीबीए) और डिमिनरलाइज्ड फ्रीज-ड्राइड बोन एलोग्राफ्ट (डीएफडीबीए) की प्रभावशीलता का तुलनात्मक मूल्यांकन, पुनः संयोजक मानव हड्डी मॉर्फोजेनेटिक प्रोटीन -2 (आरएचबीएमपी -2) के साथ: एक क्लिनिक - रेडियोग्राफिक पढाई।
2. **आमंत्रण पैराग्राफ**
   आपको एक शोध अध्ययन में भाग लेने के लिए आमंत्रित किया जा रहा है। निर्णय लेने से पहले आपके लिए यह समझना महत्वपूर्ण है कि शोध क्यों किया जा रहा है और इसमें क्या शामिल होगा। कृपया निम्नलिखित जानकारी को ध्यान से पढ़ने के लिए समय निकालें और यदि आप चाहें तो मित्रों, रिश्तेदारों और अपने इलाज करने वाले चिकित्सक/पारिवारिक चिकित्सक के साथ इस पर चर्चा करें। हमसे पूछें कि क्या कुछ ऐसा है जो स्पष्ट नहीं है या यदि आप अधिक जानकारी चाहते हैं।

3. **अध्ययन का उद्देश्य क्या है?**
   rhBMP-2 के संयोजन में अकेले DFDBA और DFDBA के साथ इंट्राबोनी दोषों के उपचार में देखे गए नैदानिक और रेडियोग्राफिक परिणामों का मूल्यांकन और तुलना करना

4. **मुझे क्यों चुना गया है?**
   आपको चुना जाता है क्योंकि आप अध्ययन के मानदंडों को पूरा करते हैं।

5. **क्या मुझे भाग लेना है?**
   यह आपको तय करना है कि भाग लेना है या नहीं। यदि आप भाग लेने का निर्णय लेते हैं, तो आपको यह सूचना पत्रक रखने के लिए दिया जाएगा और सहमति प्रपत्र पर हस्ताक्षर करने के लिए कहा जाएगा। यदि आप भाग लेने का निर्णय लेते हैं, तब भी आप किसी भी समय और बिना कोई कारण बताए वापस लेने के लिए स्वतंत्र हैं।

6. **यदि मैं भाग लेता हूँ तो मेरा क्या होगा?**
   आपको चार से पांच बार आना होगा, पहले दौरे में दांत तैयार किया जाएगा और उसके बाद हड्डी के दोष का मापन और बोन ग्राफ्ट की नियुक्ति की जाएगी और उसके बाद 6 महीने के बाद दोष का मापन किया जाएगा। एक स्वयंसेवक के रूप में, आपकी जिम्मेदारी समय पर पहुंचने की होगी।

7. मुझे क्या करना होगा?

कुछ अन्य एहतियाती उपायों के साथ आहार सेवन में कुछ बदलाव किए जाएंगे, और आपसे इसका पालन करने की अपेक्षा की जाएगी।

8. किस प्रक्रिया का परीक्षण किया जा रहा है?

ग्रुप ए में दोष स्थलों को डीमिनरलाइज्ड फ्रीज-ड्राय बोन एलोग्राफ्ट (डीएफडीबीए) के साथ ग्राफ्ट किया जाएगा। फ्लैप को बाधित टांके का उपयोग करके निकट सन्निकटन में सिल दिया जाएगा। पीरियडोंटल ड्रेसिंग लगाने से सर्जिकल साइट की सुरक्षा की जाएगी। ग्रुप बी के लिए इसी तरह की सर्जिकल प्रक्रिया की जाएगी। साइटों को रिकॉम्बिनेंट ह्यूमन बोन मॉर्फोजेनेटिक प्रोटीन -2 (आरएचबीएमपी -2) के संयोजन में डिमिनरलाइज्ड फ्रीज-ड्राय बोन एलोग्राफ्ट (डीएफडीबीए) के साथ ग्राफ्ट किया जाएगा। क्लिनिकल और रेडियोग्राफिक पुनर्मूल्यांकन के लिए और रिकॉल 3 महीने और 6 महीने में शेड्यूल किया जाएगा। प्रत्येक दौरे पर, पट्टिका नियंत्रण उपायों को सुदृढ़ किया जाएगा और यदि आवश्यक हो तो सुप्रा जिंजिवल स्केलिंग की जाएगी।

9. अध्ययन के लिए क्या हस्तक्षेप हैं?

प्री-सर्जिकल: प्रक्रिया शुरू करने से पहले सीबीसीटी प्राप्त की जाएगी सर्जिकल: इम्प्लांट साइट को एड्रेनालाईन के साथ 2% लिग्नोकेन के तहत तैयार किया जाएगा और फ्लैप की पूरी मोटाई बढ़ाई जाएगी। फिर बोन ग्राफ्ट को दोष में रखा जाएगा और हम बोन ग्राफ्ट लगाने से पहले और बाद में ऊर्ध्वाधर दोष को मापेंगे और 3 महीने और 6 महीने के बाद भी किया जाएगा। शल्य चिकित्सा के बाद: दवाएं निर्धारित की जाएंगी जैसे: एंटीबायोटिक्स, एनएसएआईडीएस।

10. भाग लेने के दुष्प्रभाव क्या हैं?

बोन ग्राफ्ट लगाने से जुड़े कुछ साइड इफेक्ट होते हैं जैसे दर्द और बेचैनी दो सप्ताह से अधिक नहीं रहती है, किसी भी बड़ी समस्या के मामले में कृपया तुरंत डॉक्टर को रिपोर्ट करें।

11. भाग लेने के संभावित नुकसान और जोखिम क्या हैं?

- किसी भी प्रणालीगत रोग के रोगी जो पीरियोडोंटल उपचार के परिणाम को प्रभावित करते हैं।
- गर्भवती और स्तनपान कराने वाली महिलाएं।
- धूम्रपान करने वाले और तंबाकू चबाने वाले।
- ऐसे मरीज जिन्होंने पिछले 3 महीनों से एंटीबायोटिक दवाओं का इस्तेमाल किया है।
- इस्तेमाल की जा रही सामग्री के लिए एक ज्ञात एलर्जी वाले विषय।

12. भाग लेने के संभावित लाभ क्या हैं?

इस अध्ययन में भाग लेने से आपको कम परेशानी में बेहतर उपचार विकल्प प्राप्त होगा। इस प्रकार के बोन ग्राफ्ट अच्छे परिणाम देंगे क्योंकि उनमें वृद्धि कारक होते हैं।

13. क्या होगा यदि नई जानकारी उपलब्ध हो जाती है?

कभी-कभी एक शोध परियोजना के दौरान, अध्ययन किए जा रहे शोध के बारे में नई जानकारी उपलब्ध हो जाती है। यदि ऐसा होता है, तो आपको इसके बारे में सूचित किया जाएगा और

अध्ययन में होने वाले परिवर्तनों के बारे में सूचित किया जाएगा। आप अध्ययन के बीच में हटने के लिए स्वतंत्र हैं। यदि आप अध्ययन जारी रखने का निर्णय लेते हैं, तो आपसे एक अद्यतन सहमति फॉर्म पर हस्ताक्षर करने के लिए कहा जा सकता है।

14. जब शोध अध्ययन बंद हो जाता है तो क्या होता है?

यदि अध्ययन निर्धारित समय से पहले समाप्त / बंद हो जाता है, तो इसका कारण रोगियों को समझाया जाएगा।

15. अगर कुछ गलत हो जाए तो क्या होगा?

बीबीडीसीओडीएस ओपीडी में क्षेत्र में विशेषज्ञता रखने वाले डॉक्टरों द्वारा स्वयंसेवकों की देखभाल की जाएगी।

16. क्या इस अध्ययन में मेरे भाग लेने को गोपनीय रखा जाएगा?

आपका नाम, पता या कोई व्यक्तिगत या अन्य जानकारी बीबीडीसीओडी के बाहर साझा नहीं की जाएगी।

17. शोध अध्ययन के परिणामों का क्या होगा?

किसी भी परिणाम/रिपोर्ट/प्रकाशन में प्रतिभागियों की पहचान का खुलासा नहीं किया जाएगा।

18. शोध का आयोजन कौन कर रहा है?

अध्ययन शोधकर्ता द्वारा आयोजित किया जाता है। बोन ग्राफ्ट का पूरा खर्च मरीज द्वारा दिया जाएगा।

19. क्या अध्ययन समाप्त होने के बाद अध्ययन के परिणाम उपलब्ध कराए जाएंगे?

यदि रोगी चाहे तो अध्ययन का परिणाम उसे उपलब्ध कराया जाएगा।

20. अध्ययन की समीक्षा किसने की है?

संस्थान के एचओडी/आईआरसी/आईईसी ने अध्ययन की समीक्षा की और उसे मंजूरी दी

21. अधिक जानकारी के लिए संपर्क करें

डॉ अंकित भदानी
पीरियोडोंटोलॉजी विभाग
पता: बनारसी दास विश्वविद्यालय, फैजाबाद रोड, आतिफ विहार, लखनऊ, यूपी। 226028
ईमेल: ankitbhadani.11@gmail.com

डॉ. लक्ष्मी बाल
संस्था की आचार समिति के सदस्य सचिव,
पता: बाबू बनारसी दास विश्वविद्यालय, फैजाबाद रोड, आतिफ विहार, लखनऊ, यूपी। 226028
ईमेल: bbdcods.iec@gmail.com

डॉसूरजपांडे (पाठक)
पीरियोडोंटोलॉजीऔरइम्प्लांटोलॉजीविभाग
बाबूबनारसीकॉलेजऑफडेंटलसाइंसेज।
लखनऊ-227105
मोब- 9628931689

डॉमोनाशर्मा (एचओडी)
पीरियोडोंटोलॉजीऔरइम्प्लांटोलॉजीविभाग
बाबूबनारसीकॉलेजऑफडेंटलसाइंसेज।
लखनऊ-227105
मोब-9984110444

bbdcods.iec@gmail.com

पं. का नाम -

पता -

ईमेल -

टेलीफोन नंबर। -

पीआई के हस्ताक्षर ……

नाम……..

तारीख……..

प्रतिभागी को सूचना पत्र की एक प्रति और हस्ताक्षरित सहमति प्रपत्र दिया जाएगा।

अध्ययन में भाग लेने के लिए धन्यवाद।

ANNEXURE – 6

Case History

DEPARTMENT OF PERIODONTICS

PATIENT'S CASE SHEET

Date: O.P.D. No.

Name: Age: Sex: Occupation

Address: Mobile No. :

CHIEF COMPLAINT(S):

HISTORY OF PRESENT ILLNESS

HISTORY OF PAST ILLNESS

A. Past Medical History

B. Past Dental History

(a) Periodontal Treatment Region

(b) **Other dental therapy**

Conservative

Prosthetics

Orthodontics

Oral Surgery

Any Other

C. Present Medical History

(a) General health

1. Bleeding Tendencies
2. Allergy
3. Cardiovascular Diseases
4. Endocrinal Diseases
5. Gastro Intestinal Diseases
6. Neurological Disorder
7. Respiratory Diseases
8. Genito Urinary Diseases
9. Hereditary/Genetic Disorder
10. Puberty/ Pregnancy/ Menopause
11. Any Infectious Disease(s)
12. Medication
13. Any other abnormality

(b) Nutritional Status: i) Well Built /Average /Poor
ii) Non Vegetarian / Vegetarian

D. PRESENT DENTAL HISTORY

(a) **Oral Hygiene Maintenance:** Brush/ Finger/ Stick / Paste/ Powder
Frequency: Once/ Twice/ Thrice
Direction

(b) **HABITS**

1.	Awareness of any Traumatising habits	Yes	No
2.	Grinding of Teeth	Yes	No
3.	Masticatory Muscle Tiredness	Morning	Evening
4.	Biting Habits	Lip/ Tongue/ Cheek/ Misc	
5.	Chewing	Betel/ Tobacco/ Mis.	
6.	Smoking	Beedi/ Cigarette/ Misc.	
7.	Mouth Breathing/ Tongue Thrusting		

CLINICAL EXAMINATION

EXTRA ORAL EXAMINATION

Face

Lips:	Competency
Skin:	Color: Normal or Palor
Neck	Swellings- Unilateral or Bilateral
Jaws:	Symmetry- Antero- Posterior relationship & movements Temporo-Mandibular Joint

INTRA ORAL EXAMINATION:

A. Soft Tissue

Labial & Buccal Mucosa:		Colour, texture
Cheek:		Colour, Stretchability, Consistency
Tongue:		Colour, Size, Mobility, Texture
Floor of the Mouth:		
Palate:	Hard:	Colour, Defect, Depth, Rougae, Tori.
	Soft:	Color, Defect
Vestibule:		
Saliva:		Flow: heavy/ diminished/ Normal Viscosity: thin/thick
Frenum/ Frenii		Number, Size, Attachment
Perio- Endo Problem		

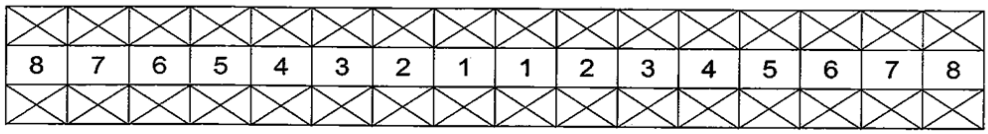
B. Gingival Status

1. Colour
2. Contour
3. Consistency
4. Surface Texture
5. Position
6. Size
7. Exudate

C. Hard Tissue

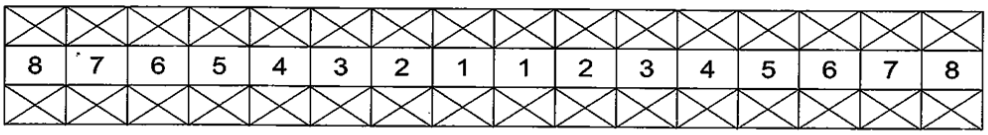
1. No. of teeth present
2. Hypersensitivity
3. Missing teeth (why, when)
4. Caries / Non-vital
5. Supernumerary
6. Proximal contact relationship
7. Plunger cusp
8. Crown size and Colour
9. Pathologic Tooth Migration
10. Mobility — Grade I / II / III
11. Hypoplasia
12. Occlusion — Angle's Classification : Class I / II / III
    Bite: Normal /Open/ Deep/Cross/Crowding
13. Retained / Impacted
14. Attrition/ Erosion/ Abrasion
15. Furcation Involvement
16. Trauma from Occlusion
17. Halitosis
18. Any dental anatomic factors
19. Calculus - Mild / Moderate / Severe
20. Stains - Mild / Moderate / Severe

19. Probing depth

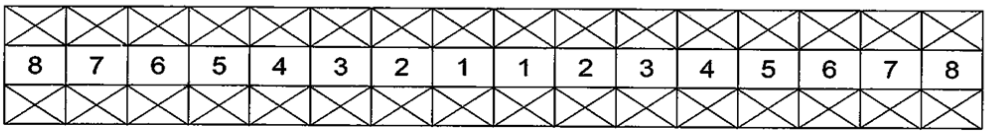


INDICES

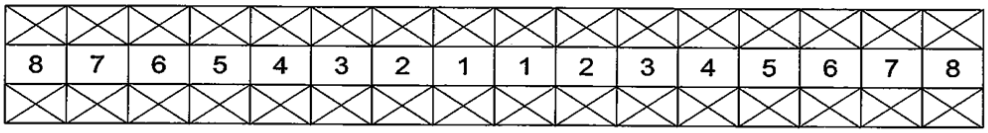
1. Plaque Index (Silness & Loe / Turesky-Gilmore-Glickman Modication of Quigley-Hein



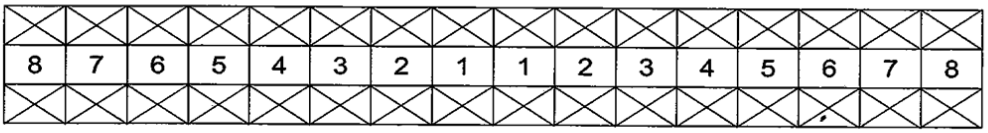
2. Gingival index (Loe & Silness / Modified Gingival Index)



3. Calculus index



4. Clinical attachment Level



DIFFERENTIAL DIAGNOSIS :

DIAGNOSIS

PROGNOSIS

TREATMENT PLAN

EMERGENCY -

PHASE I -

PHASE II -

PHASE III -

PHASE IV -

S.No.	Date	Procedure Done	Next Appointment	Staff Signature

INVESTIGATION

1. ROENTENOGRAPHIC EXAMINATION :

OPG/IOPA/BITE WING/OCCLUSAL

	DESCRIPTION	REGION
1. Lamina dura		
2. Periodontal ligament space		
3. Root form		
4. Bone loss	Vertical	
	Horizontal	
	Infra bony crater	
	Miscellaneous	
5. Periapical pathology		
6. Any other finding		

2. LAB INVESTIGATIONS

Date	Investigation	Result
	BLOOD	
	Hb%	
	RBC	
	TLC	
	DLC	
	ESR	
	Random Sugar	
	Bleeding time	
	Clotting time	
	HbS Ag Status : Positive / Negative	
	HIV Status : Positive / Negative	

DEPARTMENT OF PERIODONTICS
BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES
LUCKNOW

PROFORMA OF PATIENT'S INFORMED CONSENT

I..... son/daughter/wife of..... aged..... years, resident of..... being under the treatment of Dr..... do hereby willfully consent to the performance of a surgical procedure under local anaesthesia for the treatment of ..... (Diagnosis) upon myself / upon..... aged..... years, who is related to me as..... (for e.g. son, daughter, wife etc).

I have been informed regarding the inherent risk involved during and after the surgical procedure and that the success of the treatment cannot be guaranteed. I have signed this consent from voluntarily out of my free will without any compulsion or influence.

Date : Place :

Signature : Time :

(To be signed by parent / guardian in case of minor)

सहमति पत्र

मैं.....पुत्र/पुत्री/पत्नी.....आयु.....वर्ष

निवासी.....

मेरे दंत एवं मुख रोग का उपचार डा. ..... कर रहे हैं।

दंत एवं मसूड़े की शल्य क्रिया के लिए मुख निश्चेतना (Local anesthesia) आवश्यक है।

मुझे पूरी शल्य प्रक्रिया के दौरान होने वाले संभावित ख़तरों के बारे में ठीक से बता दिया गया है एवं उचार की सफलता के बारे में कोई निश्चितत नहीं है से भी अवगत करा दिया गया है मैं इस सहमति पत्र पर भलीभांति, बिना किसी दवाब के अपनी इच्छानुसार हस्ताक्षर कर रहा हूँ।

दिनांक हस्ताक्षर

स्थान समय

नोट : अवस्यक / नाबालिग होने की अवस्था में अभिभावक के हस्ताक्षर आवश्यक है।

ANNEXURE – 7

STATICAL ANALYSIS

The data obtained were subjected to statistical analysis using Statistical Package for the Social Sciences (SPSS Version 23; Chicago Inc., IL, USA). Data comparison was done by applying specific statistical tests to find out the statistical significance of the comparisons.

To test for effectiveness of demineralized freeze- dried bone allograft(dfdba) alone and demineralized freeze-dried bone allograft(dfdba) with recombinant human bone morphogenetic protein-2(rhbmp-2) in the treatment of intrabony defects, Shapiro Wilk tests were performed to determine the normality of the data. The test showed no significant differences and hence confirmed that the data obtained were normally distributed.

Variables were compared using mean values and standard deviation. The mean for different readings between the groups (Group A and Group B) for periodontal parameters of Plaque Index, gingival index, periodontal probing depth, clinical attachment loss and bone loss was compared using the independent 't' test. Paired t test was run to determine any difference between pre and post intervention (baseline and 6 months) for periodontal parameters. For all analysis, p value lesser than 0.05 was considered to be statistically significant.

The following formulas were employed for calculation for various parameters:

1. **Mean/Average**

Mean or Average is defined as the sum of all the given elements divided by the total number of elements

Mean = sum of elements / number of elements It is denoted by the letter X.

$$X = \frac{\Sigma X}{No.\ of observations(n)}$$

2. Standard Deviation

The standard deviation of a statistical population, a data set, or a probability distribution is the square root of its variance. Standard deviation is a widely used measure of the variability or dispersion.

It shows how much variation there is from the "Average" or Mean. It is denoted by the letter σ.

For Small samples, n<30

$$SD=\frac{\sum(X-\bar{X})^2}{n-1}$$

For Large samples, n>30

$$SD=\frac{\sum(X-\bar{X})^2}{n}$$

3. Shapiro-WilkTest

The Shapiro-Wilk test was used for testing the normality (uniformity of the distribution of the data) of the data. This approach is limited to samples between 3 and 50 elements.

The basic approach used in the Shapiro-Wilk (SW) test for normality is as follows:

A non-significant test means the sample distribution is shaped like a normal curve (uniform distribution of the values around an average value or a measure of central tendency) and parametric test are to be used.

4. **Independent‘t’ test**

The Independent Samples *t* -Test compares the means of two independent groups in order to determine whether there is statistical evidence that the associated population means are significantly different. The Independent Samples *t* -Test is a parametric test.

5. **Paired‘t’ test:**

The paired t-test, also referred to as the paired-samples t-test is used to determine whether the mean of a dependent variable is the same in two related groups i.e.., two groups of participants that are measured at two different "time points" or who undergo two different "conditions").

6. **Level of Significance (p-value)**

It is defined as the fixed probability of wrong elimination of null hypothesis when in fact it is true. In testing a given hypothesis, the maximum probability with which we would be willing to take risk is called Level of Significance of the Test.

P-value ≥ 0.05 –non-significant

P-value < 0.05 -Significant

P-value < 0.01 – Highly Significant

P-value < 0.001 - Very Highly Significant

7. **Degree of Freedom**

Degree of freedom refers to the maximum number of logically independent values, which are values that have the freedom to vary, in the data sample. Degree of freedom are commonly discussed in relation to various forms of hypothesis testing in statistics

The statistical formula to determine degrees of freedom is quite simple. It states that degrees of freedom equal number of values in data set minus 1, and it looks like this:

D f = N-1

Where N is the number of values in the data set (sample size)

8. **Bar charts:**

A bar graph is a chart that plots data using rectangular bars or columns (called bins,

can even be presented as a cylinder or a cone) that represent the total amount of observations in the data for that category. Bar charts can be displayed with vertical columns, horizontal bars, comparative bars (multiple bars to show a comparison between values), or stacked bars (bars containing multiple types of information)

Bar graphs have an x- and y-axis and can be used to showcase one, two, or many categories of data. The vertical axis of the bar graph is called the y-axis, while the bottom of a bar graph is called the x-axis. When interpreting a bar graph, the length of the bars/columns determines the value as described on the y-axis. Bar graphs are ideal for comparing two or more values or values over time.

PLAGIARISM REPORT

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