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Case Series

Effects Of Anorganic Porcine Bone And Anorganic Porcine Bone + Enamel Matrix Protein On Bone Regeneration. A Case Series Of Histological Evaluation And Comparison.

[1]Dr. Lanka Mahesh BDS, MBA, Ph.D, [2]Dr. Abhijeet Basin, MDS, [3]Dr. Sagrika Shukla, MDS (Ph.D), [4]Dr. Sumit Dubey, MDS, [5]Dr. Gaurav Mathpal MDS, Ph.D, [6]Dr. Nilesh Rathi, [7]Dr. Nikita Gulati, BDS, MDS

[1]*Adjunct prof. dept. of Periodontology and Implantology.Dr. D.Y. Patil Dental College and hospital Vidyapeeth,Pimpri,Pune,India.*
[2]*Senior Lecturer,Department of Periodontology and Implantology Bhabha College of Dental Sciences,Bhopal, India.*
[3]*Reader, Dept. of Periodontology and Implantology,SGT Dental College,SGT University, Budhera, Gurgaon,India.*
[4]*Reader, Dept of Prosthodontics,DJ Dental college and hospital,Modinagar, India.*
[5]*Senior Consultant, (Prosthodontics)Panacea Clinic R. New Delhi.*
6Prof and head,Dept. of pediatric and preventive dentistry,Dr. D.Y. Patil Dental College and hospital,Vidyapeeth,Pimpri, Pune.
[7]*Reader, Dept of Oral and Maxillofacial Pathology and Microbiology,ITS-CDSR, Muradnagar,India.*

Abstract

objective: to histologically analyse regeneration between anorganic porcine bone (MinerOss® XP) and anorganic porcine bone (MinerOss® XP) + enamel matrix protein (Emdogain®)
Materials and method: In total 20 patients were enrolled for the study wherein; 10 patients (group A) were grafted with anorganic porcine bone (MinerOss® XP) and 10 (group B) with anorganic porcine bone + Emdogain®. After 5 months, at the time of implant placement 3 mm bone cores were taken with trephine for histological evaluation. Osteotomy was prepared and implants were placed.
Results: The group A, treated with MinerOss® XP alone predominantly exhibited mature bony trabeculae interspersed with residual graft material and group B, treated with anorganic porcine bone (MinerOss® XP) + enamel matrix protein (Emdogain®) revealed well-formed trabecular bone exhibiting various stages of mineralization.
Conclusion: Overall, both graft groups demonstrated comparable histological bone quality, indicating effective bone regeneration potential.

Keywords: bone graft, socket preservation, regeneration, xenograft, Emdogain®.

INTRODUCTION

Bovine xenograft was first used for bone augmentation in 1920s, since then, science of guided bone regeneration (GBR) is ever evolving. Today, bone graft substitutes play a huge role from bone augmentation to imparting implant stability. Bone augmentation or bone grafting starts with tooth extraction in the form of socket preservation, which is one of the most important step in implant's proper placement and function. It is an established fact that tooth extraction results in bone resorption, maximum of which occurs in the first 6 months unsuitable for implant placement1-3. Mahesh et al4 in a case series showed that Clinically and radiographically (CBCT) by 5th week massive bone resorption occurs. Schropp et al5 stated that 2/3rd of the hard and soft tissue changes occur in the first 3 months and 50% of crestal width is lost in a 12-month period, 2/3 of which occurred in just the first 12 weeks. Hence, bone graft substitutes become paramount in the treatment of socket preservation as it provides adequate amount of bone for implant placement and preserve bone from further loss at the future implant site6.

By definition, Socket preservation (or socket seal surgery (SSS) as previously known), is a prophylactic procedure that aims to minimize alveolar ridge atrophy after tooth extraction7. And in literature many techniques have been applied such as atraumatic extraction, drilling through the residual roots in

***Corresponding Author:** Dr.Sagrika Shukla,MDS,Ph.D.* Reader, dept of Periodontology,SGT dental college and hospital,Budhera, Haryana.
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order to preserve bone8, use of barrier membranes to cover the socket7, socket-plug technique and when it comes to materials, all types of bone graft substitutes have been used with varying degree of success8-10. The choice of material is vast autogenous bone, various types of xenografts, allografts and alloplastic bone grafts all can be and have been used for socket preservation with successful results. The choice of material lies upon many factors and it most importantly depends upon the properties of the graft used and clinician's desired outcome. For instance, a bovine xenograft takes prolonged time for absorption, its this property is advantageous in case there is prolonged time between grafting and osteotomy, however the same cannot be said for autografts. According to Klinge and colleagues11 bovine xenografts provide an ideal scaffold for new bone formation and supports osteoblastic cell attachment and proliferation when used in rabbits. However, histological studies have revealed the presence of remnants of amorphous graft particles even after several months following its implantation in vivo. There are many other advantages of xenografts which makes their choice more prominent as compared to other types of grafts. One such graft used in this study is MinerOss® XP (BioHorizons, USA) which is a highly porous anorganic porcine bone mineral matrix. Its increased porosity provides increased osteoconductivity and adequate space for new bone deposition.

In the current study MinerOss® XP high osteoconductivity property which has been explored along with Emdogain® which is a is a mixture of enamel matrix derivatives (EMDs) used for regeneration.

MATERIAL AND METHOD

The study was ethically conducted at a private dental office wherein, healthy patients were enrolled who were willing for implant treatment under the following inclusion and exclusion criteria:

Inclusion criteria:

- Above 18 years of age
- Medically healthy without any disease, or controlled medical conditions such as Diabetes.
- Good oral hygiene
- Low or no smoking
- Compliant patient.

Exclusion criteria:

- Non-compliant patient
- Medically compromised
- Pregnant or breast-feeding mothers
- At the site of Active infection
- Patient on immunosuppressants
- Addiction of alcohol or drug abuse
- Psychiatric disorders – patients inability to comprehend instructions.

In total 20 patients were enrolled for the study, patients were divided into two groups

Group A- 10 patients grafted with MinerOss® XP,

Group B - 10 patients grafted with MinerOss® XP + Emdogain® (Straumann, Basel, Switzerland).

After tooth extraction (fractured or grossly decayed), the site was grafted with MinerOss® XP, or with MinerOss® XP + Emdogain®. The selection of grafted material was non-randomised, which was mostly dependent upon the affordability of the grafting solutions. A colaplug was placed and it was sutured. After a period of 5 months the site was re-visited for implant placement wherein, before starting with osteotomy, a bone core of 3mm was procured with trephine for histological evaluation followed by 5/10 Bioner implants placement, suturing and post-operative instructions.

All 20 patients showed uneventful healing, the graft and implant take-up was clinically well without any signs of allergies, rejection or implant failure, although mild discomfort was reported by a few patients related to surgery.

HISTOLOGY

The 3 mm trephine core biopsies, along with the overlying soft tissue, were immediately fixed in 10% neutral buffered formalin for 24 to 48 hours. Following fixation, the hard tissue specimens were decalcified in 10% ethylenediaminetetraacetic acid (EDTA, pH 7.4) using a gentle decalcification protocol. Both hard and soft tissue specimens were then subjected to routine tissue processing, including dehydration, clearing, and paraffin wax infiltration. Paraffin-embedded tissue blocks were prepared and sectioned at a thickness of 3 µm. The sections were stained using Hematoxylin and Eosin (H&E) and examined under an Olympus BX53 research microscope. Representative low- and high-power digital photomicrographs were captured using an Olympus EPL3 digital camera.

RESULTS

Specimens treated with MinerOss® XP alone predominantly exhibited mature bony trabeculae interspersed with residual graft material. The trabeculae showed well-defined osteoblastic rimming and entrapped osteocytes. **Figure 1-4**.

Figure 1. H and E staining at 4x magnification showing dense stroma with bony trabeculae.

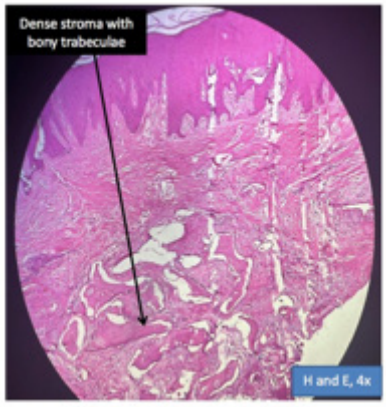


Figure 2. H and E staining at 10-40x magnification Residual membranous graft material entrapped between bone trabeculae.

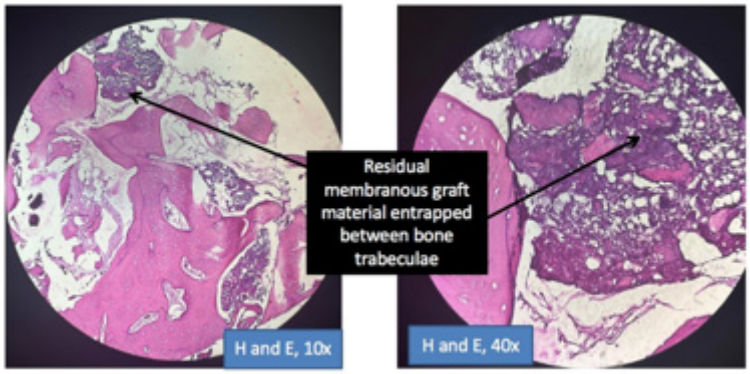


Figure 3. H and E staining at 40x magnification.

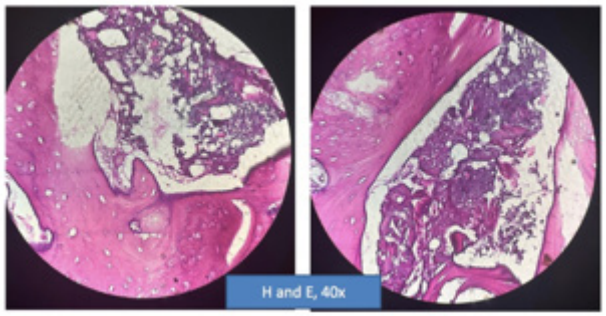
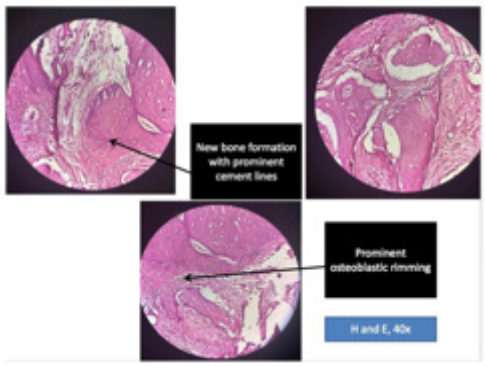


Figure 4. H and E staining at 40x magnification showing new bone formation with prominent cement lines.



In contrast, Histological examination of the H&E-stained sections from the Emdogain® + MinerOss® XP bone tissue cores revealed well-formed trabecular bone exhibiting various stages of mineralization. At the graft–new bone interface, numerous immature bony trabeculae with abundant entrapped osteocytes, osteoblastic rimming, and vascular connective tissue were observed. Focal areas demonstrated new bone formation interspersed with residual graft material. **Figure 5-8**
In the majority of the bone cores, osteocytes entrapped within lacunae and bone-lining cells on the mature trabeculae displayed distinct lamellations, along with prominent resting and reversal lines. The coexistence of woven and mature bone suggested favourable osteoinductive and osteoconductive properties.

Figure 5. H and E staining at 10x magnification interspersed bony trabeculae with graft material.

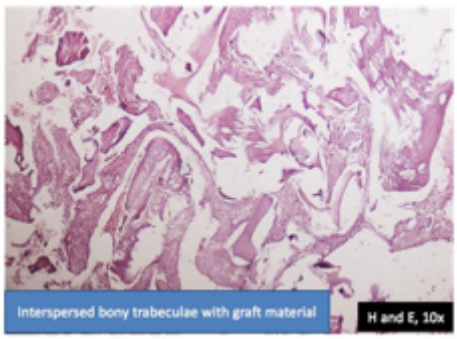


Figure 6. H and E staining at 10x magnification showing new bone formation.

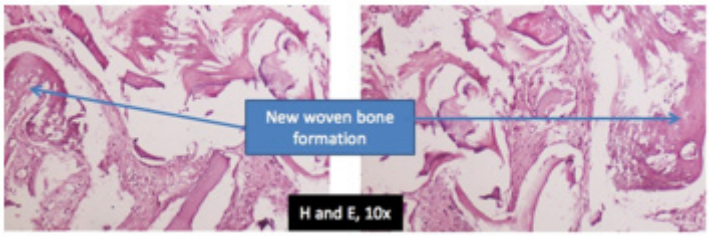


Figure 7. H and E staining at 40x magnification showing residual graft material.

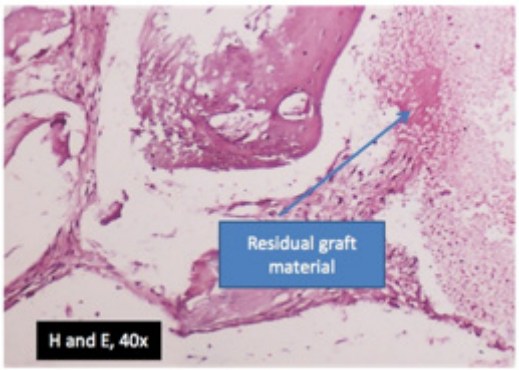
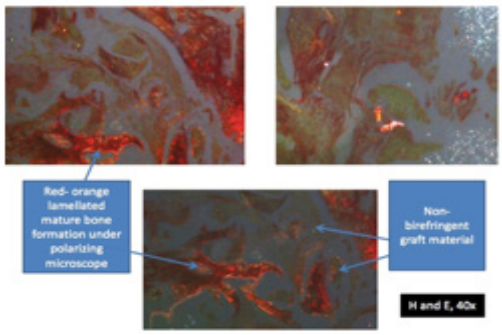


Figure 8. H and E staining at 40x magnification showing red- orange lamellated mature bone formation under polarizing microscope.



DISCUSSION

As aforementioned, choice of graft does play a role in regeneration of the desired tissue. The gold standard is autogenous bone but its limited availability, second surgical site and fast resorption rate does not make it an ideal bone graft substitute in case of socket preservation. Socket preservation is a method to 'preserve' bone for future implant placement. Hence the choice of graft should be able to keep the bony dimensions stable or without resorption for a longer period of usual time period and the graft which fits the option is xenograft. There are reports where a xenograft has been found at the surgical site even after 20 years of its placement. Mordenfeld et al12 showed presence of deproteinized bovine bone particles which even after 10 years had not biodegraded. Mordenfeld et al13 in histological studies detected particles of bovine bone with no significant size change at 11 years. Ayna et al14 showed the presence of residual bovine bone particles in humans after 14 years. Traini et al15 reported residual particles of anorganic bovine bone after 20 years.

Graft particles present upto such prolong time are 'graft remanent' as any grafted site does undergo continuous modelling and re-modelling4. However, Mordenfeld et al13 noticed presence of multinucleated giant cells (MGC) around un-resorbed graft. Literature states that any residual graft or unabsorbed graft will show presence of MGC as commonly seen in foreign-body reactions. Yet more recently these cells have been shown as possible contributors to tissue regeneration16. In the aforementioned studies12-15 histologically and clinically bone volumes have been shown to be stable without any patient discomfort. Currently, how these MGCs are capable of maintaining long-term bone volumes without any obvious reactions is unknown and needs further research. Additionally, implants placed in 100% xenograft grafted sites show higher survival rate (95.6%) as compared to those grafted in 100% autogenous bone (88.0%) or grafted with allografts (81.0%)17. MinerOss® XP being a xenograft helps in augmentation of variety of bony defects such as periodontal defects, ridge augmentation, sinus augmentation, 3-walled and 4-walled defects, block grafting with successful results.

In spite of vast literature available on efficiency of a xenograft being regenerative in nature the authors grafted MinerOss® XP + Emdogain® (Strau-mann, Basel, Switzerland). The rationale behind this was that Emdogain® is an osteopromotive agent for regeneration. It is a mixture of enamel matrix derivatives (EMDs). EMD is enamel maxtrix extract, derived from porcine teeth which contains various proteins out if which 90% is amelogenins, which aid in perio-dontium attachment at the time of tooth development18. It also contains other components such as ameloblastin, tuftelin, enamelin, and amelotin19. EMD is one component which since its FDA approval in 1996 has been extensively studied and used for periodontal regeneration successfully and safely. And one of the most common commercially available EMD is Emdogain® (Strau-mann, Basel, Switzerland) with about 15 years of supportive clinical data, it enhances the proliferation of osteoblasts20, promotes cell differentiation21 and stimulating migration and viability of osteoblasts22. At the molecular level, it upregulates Runx2 and Osterixtranscription factors, dentin sialophosphoprotein, dentin matrixprotein 1, and osteopontin RNA in human dentin pulp stem cells23,24.

Through the current study it is safe to state that MinerOss® XP aids in regeneration, so far according to authors knowledge there is no study which can state presence of residual graft after few years of its placement. Also, our results showed that both the groups demonstrated comparable histological bone quality, indicating effective bone regeneration potential. This proves that with a superior graft such as MinerOss® XP, addition of any other regenerative material is not needed.

CONCLUSION

Overall, both graft groups demonstrated comparable histological bone quality, indicating effective bone regeneration potential.

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Conflict of interest
none

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