



Case reports

# LOCALIZED BONE REGENERATION WITH PORCINE BONE: 18-YEAR FOLLOW-UP

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#### **ABSTRACT**

The presence of localised bone defects in the alveolar processes can prevent the use of implants, due to insufficient bone volume for osseointegration. Localised bone resorption can also prevent a good aesthetic result due to poor soft tissue support. Autologous bone grafting is considered the gold standard, but it has disadvantages for the patient, such as a donor site and the risk of morbidity. The use of alloplastic, allogenic, or xenogenic grafts has therefore become a very attractive alternative. A recent study conducted on the maxilla of rabbits has shown that porcine bone grafting exhibits strong osteoconductive properties and, over time, is remodelled and replaced with new bone. The aim of this study is to describe a technique designed to reconstruct a bone volume suitable for implant placement and, therefore, capable of supporting soft tissues in order to achieve an adequate aesthetic result with an 18-year follow-up of prosthetic loading. Bone augmentation was also evaluated from a histological point of view at the time of dental implant placement.

**KEYWORDS**: dental implants, bone regeneration, xenogeneic grafts, porcine bone

## INTRODUCTION

Adequate bone volume is a fundamental prerequisite for successful implant placement, ensuring functional, aesthetic, and prosthetic rehabilitation. Clinical and histological studies in both animals and humans have demonstrated that spontaneous healing of post-extraction sockets initiates a physiological process resulting in bone remodeling and resorption, which is more pronounced on the buccal side than on the palatal or lingual aspects (1-3).

In 1996, Buser et al. (4) introduced the concept of Guided Bone Regeneration (GBR) to address the loss of alveolar bone. The aim of GBR is to promote new bone formation in ridge defects, either prior to or simultaneously with implant placement. GBR relies on creating a favorable environment that supports the natural regeneration of bone tissue (5-7). Critical factors in establishing this environment include stabilization of the blood clot, prevention of acute inflammation due to bacterial infection, and the creation and maintenance of a space filled with the clot (8-9).

Both resorbable and non-resorbable barrier membranes have been successfully employed in bone regeneration for many years. Their primary function is to exclude soft tissue cells from the mucosa, thereby allowing osteogenic cells from surrounding tissues to repopulate the defect. However, most membranes alone are insufficient to maintain a stable

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space conducive to bone formation. While autologous bone grafts, in block or particulate form, remain the gold standard for ridge augmentation (10-11), alternative grafting materials, including xenografts (12-13), alloplastic grafts (14-15), and allografts (16), have been extensively studied. The use of alternative materials is driven by the desire to eliminate the need for a second surgical site, thereby reducing procedural invasiveness and postoperative morbidity. A recent study on rabbit maxillae confirmed that porcine-derived bone grafts exhibit strong osteoconductive properties and are gradually resorbed and replaced by new bone over time (17).

The aim of this study is to report on the clinical, radiographic, and histological outcomes observed following the use of porcine bone granules (Osteobiol Gen-Os, Tecnoss, Italy), mixed with collagen gel and covered with a thin cortical lamina, six months after the regenerative procedure. Long-term clinical and radiographic outcomes are also assessed after 18 years of prosthetic loading (18).

### MATERIALS AND METHODS

In February 2007, two patients were recruited who had a horizontal and, in part, vertical bone defect in the upper first premolar. Since both bone defects were quite extensive, it was decided to carry out the regeneration procedure in two stages: bone regeneration, followed by a six-month waiting period, and then implant insertion with a healing period of at least four months before reopening and abutment placement. The area to be regenerated was filled with a mixture of collagen gel (OsteoBiol Gel 0, Tecnoss, Italy) mixed with collagenated bone of porcine origin (OsteoBiol Gen-Os, Tecnoss, Italy). This particular mixture has allowed for better control of the grafting, thanks to the greater malleability of the product obtained and its particular adhesiveness.

The main purpose of the proposed protocol was to maintain adequate space for new bone regeneration, which was achieved by using a support screw beneath a cortical bone plate (OsteoBiol Soft Cortical Lamina, Tecnoss, Italy). The latter was stabilised on the vestibular side with two mini osteosynthesis screws (length 5 mm, diameter 1.2 mm; Graftek fixation screws, Roen), while on the palatal side, it was positioned below the mucoperiosteal flap.

After six months, at the end of the healing period, a bimodal surface implant with positive tolerance geometry and a taper of approximately 1° (Neoss Italia) was inserted (19). After implant placement, at the time of reopening and before positioning the final abutment, the ISQ (implant stability quotient, Osstell Mentor) value was measured, i.e., the resonance frequency value, which is related to the stiffness of the bone-titanium interface.

The implant site was prepared using a Trephine drill (Maillefer) with an internal diameter of 2 mm and an external diameter of 3 mm. The drill, containing the extracted bone, was immersed in a 4% buffered formaldehyde solution and sent to the Department of Biomaterials at the Institute of Surgical Sciences (Sahlgrenska Academy, Gothenburg University, Sweden) for histological evaluation. The samples were then dehydrated with successive steps in different alcohol gradients and subsequently embedded in photopolymerisable resin. Sections of approximately 10-15 µm were cut using a saw and a grinder, stained with toluidine blue, and observed under a microscope.

#### First case

A 56-year-old female patient with edentulism in zone 1.4 presents with both vertical and horizontal bone defects. Consequently, it is decided to perform bone defect reconstruction using GBR prior to implant placement (Fig. 1).

Under local anaesthesia (2% lidocaine with 1:80,000 epinephrine), a full-thickness vestibular flap is designed in order to expose the bone defect completely. After carefully cleaning the bone surface of any periosteal residue, the vestibular cortical bone of the recipient site was perforated with a ball bur to induce bleeding and promote the incorporation and vascularisation of the graft material (Fig. 2).



**Fig. 1.** *Pre-operative radiograph.* 



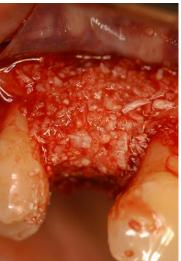
**Fig. 2.** *Intraoperative view of the bone defect.* 

A support screw is inserted on the occlusal side of the recipient site in order to create a tenting effect and thus prevent the barrier membrane from collapsing. The site is then filled with OsteoBiol Gen-Os mixed with collagen gel (OsteoBiol Gel 0) to make the product denser and stickier, thereby facilitating its positioning (Fig. 3).

A thin cortical lamina (OsteoBiol Lamina Soft) is moulded over the graft material and stabilised to the bone using two fixation screws on the vestibular side apically to the defect itself, while on the palatal side, the lamina is adapted to the bone surface below the mucoperiosteal flap (Fig. 4).



**Fig. 4.** Placement of membrane with fixation screws.



**Fig. 3.** Defect filled with Osteobiol graft material.

Before suturing, a fenestration of the periosteum is performed at the base of the vestibular flap in order to achieve a tension-free adaptation of the wound margins. The wound is closed with horizontal mattress sutures and separate stitches. The patient was called back weekly for follow-up visits.

At the end of the third week, partial exposure of the osteosynthesis screw head was noted, which was nevertheless maintained. The patient was advised to apply chlorhexidine gel daily. After six months, at the surgical follow-up, no residual lamina or biomaterial granules were detected, but rather compact and well-vascularised new bone formation. The partial exposure of the upper head of the fixation screw indicates that there has been minimal vertical resorption of the graft material (Fig. 5).

The increase in crest height was quantifiable as 5 mm horizontally and 4 mm vertically. Once the screw was removed, a small portion of the regenerated bone was taken for histological examination using a 3 mm external diameter drill bit (Fig. 6).



**Fig. 5.** Bone regeneration observed at surgical re-entry, six months postoperatively.



**Fig. 6.** Histological result from the implant insertion site: Biopsy performed at 5 months shows mature bone. Bone substitute particles (\*) are barely distinguishable from newly formed bone. (Scale bar =  $200 \mu m$ ).

Subsequently, the preparation of the implant socket was completed, and a 13 mm long, 4 mm diameter implant (Neoss Ltd, Harrogate, UK) was inserted, achieving good primary stability (ISQ 71). A 3 mm high transgingival healing cap made of PEEK (polyetheretherketone) is immediately inserted to manage the soft tissues and avoid further surgery.

After a five-month healing period, implant stability was reassessed (ISQ 78); the X-ray examination showed that the implant was well integrated into the basal bone. Subsequently, the implant was restored with a gold-ceramic crown (Fig. 7, 8).



Fig. 7. Clinical view at prosthetic delivery.



Fig. 8. Radiograph at the time of prosthetic delivery.

#### Second case

A 38-year-old female patient presented to our clinic with traumatic avulsion of tooth 2.4 (Fig. 9). The residual area showed a severe horizontal and vertical defect and, in particular, a loss of attachment of approximately 7 mm, mesially to tooth 2.5. As per protocol, the area was anaesthetised and skeletonised and, after drilling holes in the bone to be grafted, a regeneration screw (Memphix, Straumann) was placed, with a smooth part of 5 mm outside the bone (Fig. 10). A soft cortical lamina (OsteoBiol Lamina, Tecnoss) was moulded and fixed with two mini screws (5 mm long and 1.2 mm in diameter, Graftek fixation screws, Roen) vestibularly to the defect. After the cortical lamina had been properly adjusted, the defect was filled with a mixture of collagen and bone (OsteoBiol Gen-Os, Tecnoss) and then covered with the cortical lamina (Fig. 11).



Fig. 9. Pre-operative radiograph.



**Fig. 10.** Placement of a tenting screw to facilitate space maintenance during regeneration.

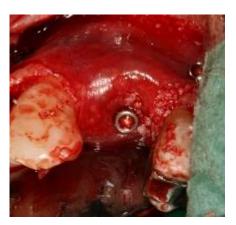


Fig. 11. Cortical bone lamina fixed to the buccal side, covering the graft.

The release of the flap with a periosteal incision and the use of mattress sutures allowed the flap to be closed and the graft to be covered throughout the healing period. After a seven-month waiting period, with no symptoms or signs of inflammation or infection, the flap was reopened, the screws were removed, and the implant site was prepared with a Trephine drill.

For histological evaluations, bone sampling is performed. The site was refined using a 3.2 mm diameter drill. A Neoss implant (length 13 mm and diameter 4 mm) was inserted, achieving excellent primary stability (ISQ 77). Bone measurements showed an increase of 5 mm vertically and 4 mm horizontally; in addition, mesially to the second premolar, a localised attachment gain was noted (Fig. 12).



After five months, the site was reopened, a zirconium abutment and a temporary crown were immediately placed, and an increase in resonance frequency values (ISQ 81) was measured. After four months, a permanent zirconium and ceramic crown was cemented. The final intraoral X-ray confirms the increase in bone growth (Fig. 13, 14).

Histological examination of the bone biopsy from the site shows well-incorporated bone substitute particles and dense bone (Fig. 15).

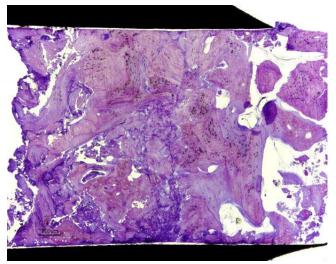
**Fig. 12.** Surgical re-entry at 7 months: a horizontal and vertical bone gain of 5 mm is noted, along with improved attachment mesial to tooth 25.



**Fig. 13.** Clinical view of the zirconia crown one-year post-grafting.



Fig. 14. Radiograph 18 months after grafting.



**Fig. 15.** Histological result: presence of dense mature bone with visible bone substitute particles (\*). (Scale bar = 200  $\mu$ m).

#### DISCUSSION

The original aim of the study was to evaluate the clinical, radiological and histological results obtained six months after regenerative surgery using a mixture of collagen gel (OsteoBiol Gel 0, Tecnoss) combined with collagenated bone of porcine origin (OsteoBiol Gen-Os, Tecnoss) as a filling material in combination with a cortical bone lamella (OsteoBiol Lamina Corticale Soft, Tecnoss). In both cases, there was an increase in bone ridges, both vertically and horizontally, of approximately 4-5 mm and an increase in the volume of local soft tissue, which allowed the implants to be inserted in an appropriate position, satisfying both functional and aesthetic criteria. There were no complications, except in one of the two cases, where, after approximately three weeks, partial exposure of the head of the osteosynthesis screw occurred. However, this did not cause any inflammatory or infectious reaction and did not affect bone healing. These data demonstrate the optimal biocompatibility of the materials used.

A comparison of the ISQ values at the time of implant placement and at the time of abutment placement after ten months showed a significant increase in both cases (first case: from 71 to 78; second case: from 77 to 81), indicating that a clear process of bone remodelling had taken place, leading to greater densification and maturation of the new bone tissue.

The results of histological examinations performed six months after GBR showed the presence of mature bone. Furthermore, the porcine bone particles were well incorporated and difficult to distinguish from native bone. In some areas, remodelling was observed with partial resorption of the particles and the formation of new bone. Particles without bone contact could occasionally be seen in the deepest areas of the two biopsies.

Similar data were also reported by Nannmark and Sennerby (17) in an animal model. The authors evaluated bone tissue responses to PCPB, with or without collagen gel, covered with a collagen membrane (OsteoBiol Evolution, Tecnoss). Histological examinations performed at eight weeks showed active resorption of the materials, the presence of mature bone and revascularisation of the mineralised part and soft tissue, and finally that the collagen membrane was undergoing active degradation.

After 18 years of prosthetic loading, during which the two patients examined did not experience any complications, radiological examination showed that the crestal bone levels remained unchanged. (Fig. 16, 17).



**Fig. 16.** Radiographic follow-up of Case One at 18 years of prosthetic loading.



**Fig. 17.** Radiographic follow-up of Case Two at 18 years, showing excellent stability of the bone tissues. Over time, a post-extraction implant was placed at site 25 with maxillary sinus lift via a crestal approach using the "Rialto" technique.

### **CONCLUSIONS**

This study has provided clinical, histological, and radiological evidence in humans of the regenerative potential of the combination of the three materials used (OsteoBiol Gel 0, OsteoBiol Gen-Os, OsteoBiol Soft Cortical Lamina, Tecnoss) after 18 years of functional loading.

# REFERENCES

- 1. Amler MH, Johnson PL, Salman I. Histological and histochemical investigation of human alveolar socket healing in undisturbed extraction wounds. *The Journal of the American Dental Association*. 1960;61(1):32-44. doi:https://doi.org/10.14219/jada.archive.1960.0152
- 2. Lekovic V, Kenney EB, Weinlaender M, et al. A bone regenerative approach to alveolar ridge maintenance following tooth extraction. Report of 10 cases. *Journal of Periodontology*. 1997;68(6):563-570. doi:https://doi.org/10.1902/jop.1997.68.6.563
- 3. Araújo MG, Lindhe J. Dimensional ridge alterations following tooth extraction. An experimental study in the dog. *Journal of Clinical Periodontology*. 2005;32(2):212-218. doi:https://doi.org/10.1111/j.1600-051X.2005.00642.x
- 4. Buser D, Dula K, Hirt HP, Schenk RK. Lateral ridge augmentation using autografts and barrier membranes: A clinical study with 40 partially edentulous patients. *Journal of Oral and Maxillofacial Surgery*. 1996;54(4):420-432. doi:https://doi.org/10.1016/s0278-2391(96)90113-5
- 5. Buser D, Dula K, Belser U, Hirt HP, Berthold H. Localized ridge augmentation using guided bone regeneration. 1. Surgical procedure in the maxilla. *The International journal of periodontics & restorative dentistry*. 1993;13(1):29-45.
- Dahlin C, Andersson L, Linde A. Bone augmentation at fenestrated implants by an osteopromotive membrane technique. A controlled clinical study. *Clinical Oral Implants Research*. 1991;2(4):159-165. doi:https://doi.org/10.1034/j.1600-0501.1991.020401.x
- 7. Schenk RK, Buser D, Hardwick WR, Dahlin C. Healing pattern of bone regeneration in membrane-protected defects: a histologic study in the canine mandible. *PubMed*. 1994;9(1):13-29.

8. Scantlebury TV. 1982-1992: A Decade of Technology Development for Guided Tissue Regeneration. *Journal of Periodontology*. 1993;64(11s):1129-1137. doi:https://doi.org/10.1902/jop.1993.64.11s.1129

- 9. Hardwick R. Membrane design criteria for guided bone regeneration of the alveolar ridge. *Chicago, Quintessence*. Published online January 1, 1994:101-135.
- 10. Misch CM, Misch CE, Resnik RR, Ismail YH. Reconstruction of maxillary alveolar defects with mandibular symphysis grafts for dental implants: a preliminary procedural report. *PubMed*. 1992;7(3):360-366.
- 11. Simion M, Jovanovic SA, P Trisi, Scarano A, A Piattelli. Vertical ridge augmentation around dental implants using a membrane technique and autogenous bone or allografts in humans. *Int J Period Rest Dent* . 1998;18(1):8-23.
- 12. Proussaefs P, Lozada J, Kleinman A, Rohrer MD. The use of ramus autogenous block grafts for vertical alveolar ridge augmentation and implant placement: a pilot study. *PubMed*. 2002;17(2):238-248.
- 13. Skoglund A, Hising P, Young C. A clinical and histologic examination in humans of the osseous response to implanted natural bone mineral. *PubMed*. 1997;12(2):194-199.
- 14. Kent JT, Quinn JV, Zide MF, María L, Boyne P. Alveolar ridge augmentation using nonresorbable hydroxylapatite with or without autogenous cancellous bone. *Journal of Oral and Maxillofacial Surgery*. 1983;41(10):629-642. doi:https://doi.org/10.1016/0278-2391(83)90016-2
- 15. Holmes R, Mooney V, Bucholz R, Tencer A. A Coralline Hydroxyapatite Bone Graft Substitute. *Clinical Orthopaedics and Related Research*. 1984;&NA;(188):252???262. doi:https://doi.org/10.1097/00003086-198409000-00036
- 16. Nevins M, Mellonig JT, Clem DS, Reiser GM, Buser DA. Implants in regenerated bone: long-term survival. *Int J Period Rest Dent*. 1998;18(1):34-45.
- 17. Nannmark U, Sennerby L. The Bone Tissue Responses to Prehydrated and Collagenated Cortico-Cancellous Porcine Bone Grafts: A Study in Rabbit Maxillary Defects. *Clinical Implant Dentistry and Related Research*. 2008;0(0):080131095245125-??? doi:https://doi.org/10.1111/j.1708-8208.2007.00080.x
- 18. Sennerby L, Meredith N. Implant stability measurements using resonance frequency analysis: biological and biomechanical aspects and clinical implications. *Periodontology* 2000. 2008;47(1):51-66. doi:https://doi.org/10.1111/j.1600-0757.2008.00267.x
- 19. Volpe S, Verrocchi D, Andersson P, Gottlow J, Sennerby L. Comparison of early bacterial colonization of PEEK and titanium healing abutments using real%time PCR. *Applied Osseointegration Res.* 2008;6:54-56.