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Autologous bone transfer is regarded as the gold standard for ridge augmentation before dental implantation, especially in severe bony defects caused by tumor resection or atrophy. In addition to the advantages of autologous bone, transplantation has several disadvantages, such as secondary operation, increased morbidity and pain. The present study reports, for the first time, a combination of a xenogeneic bone substitute (BO) with platelet-rich fibrin (PRF), which is a fully autologous blood concentrate derived from the patient’s own peripheral blood by centrifugation. Solid A-PRF and liquid i-PRF together with an individualized 3-D planned titanium mesh were used for reconstruction of a severe tumor-related bony defect within the mandible of a former head and neck cancer patient. The BO enriched with regenerative components from PRF allowed the reconstruction of the mandibular resective defect under the 3-D mesh without autologous bone transplantation. Complete rehabilitation and restoration of the patient’s oral function were achieved. Histological analysis of extracted bone biopsies confirmed that the new bone within the augmented region originated from the residual bone. Within the limitations of the presented case, the applied concept appears to be a promising approach to increase the regenerative capacity of a bone substitute material, as well as decrease the demand for autologous bone transplantation, even in cases in which autologous bone is considered the golden standard. PRF can be considered a reliable source for increasing the biological capacities of bone substitute materials.

Key Words: vertical augmentation, Bio-Oss, platelet-rich fibrin (PRF), titanium mesh, head and neck cancer, LSCC

INTRODUCTION

Numerous investigations and studies within past years have demonstrated that placement of dental implants is a reliable, long-term, stable method to restore oral function, which includes articulation, mastication, and aesthetics. In addition to the technical requirements of placing implants, it was further shown that a sufficient amount and quality of bone and soft tissue in the prospective implant site are of striking importance in achieving osseointegration, long-term stability and peri-implant health. Conventional augmentation procedures such as guided bone regeneration (GBR) allow simultaneous augmentation for implant placement in cases of modest horizontal or vertical bone deficits. Usually, the applied bone substitute materials serve as an osteoconductive scaffold for ingrowth of newly formed bone from surrounding native bone. A collagen membrane is used to function as a barrier to prevent the ingrowth of soft tissue in the augmentation bed. However, in complex cases—such as after tumor resection or in cases of severe atrophy—these augmentation techniques are stretched to their limits. It is postulated that in extensive augmentation procedures, a sufficient implantation bed can be generated only by means of autologous bone transplantation. However, these procedures are associated with a remarkable burden to the patient, including a secondary operative site, pain, and wound healing disorders at the donor site.

Due to the aforementioned disadvantages of autologous bone transfers, numerous bone substitute materials of different origins have been widely investigated. These materials are all similar in that they do not possess osteoinductive properties and therefore are limited to osteoconductive properties. Previous investigations of our workgroup have demonstrated that the origin of bone substitute materials has a major influence on tissue reaction within the host organism. In different in vivo and clinical trials, it was shown that the physicochemical properties of bone substitute materials have an impact, especially regarding vascularization and the evoked cellular inflammatory pattern within the implantation bed (ie, induction of multinucleated giant cells, which influences biomaterial degradation).

A promising approach to enhance the regenerative potential of bone substitute materials is the application of an

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autologous blood concentrate system, otherwise known as platelet-rich-fibrin (PRF). This system is obtained by the centrifugation of a patient’s own peripheral blood without additional anticoagulants, resulting in solid and fluid PRF-matrices. Recently, the so-called low-speed centrifugation concept (LSCC) showed that the reduction of the applied relative centrifugation force (RCF) during centrifugation results in significant enrichment of these matrices with leukocytes, platelets, and their growth factors. Based on the LSCC, optimized protocols for the solid advanced PRF+ (A-PRF+) and the fluid injectable PRF (i-PRF) were shown to include a significantly higher number of platelets and leukocytes while possessing a high bioactivity in terms of the release of different growth factors. The included autologous components may promote acceleration of wound healing and tissue regeneration without adding further external substances. For the solid PRF-matrix, blood components—such as B- and T-lymphocytes, monocytes, stem cells, granulocytes, and growth factors—are embedded in a fibrin clot. Within the fluid PRF-matrix, however, all the aforementioned cells together with their growth factors were concentrated in a 1- to 1.5-mL fluid volume.

For the first time, this presented case reports a combination of a xenogeneic bone substitute (BO) with solid A-PRF+ and liquid i-PRF for reconstruction of a severe tumor-related bony defect in the mandible of a former head and neck cancer patient. It was postulated that the use of a well-processed and well-investigated xenogeneic bone substitute material (such as the one we choose) together with both aforementioned PRF components may sufficiently regenerate bone without the need of autologous bone.

**Presented Case**

The presented case was a 61-year-old female patient affected by squamous cell carcinoma (pT2N0M0) in the anterior floor of the mouth, treated by tumor resection including a block-type resection of the mandible along with bilateral neck dissection (see Figures 2 and 3). Sixteen months after tumor therapy as well as regular radiological and clinical tumor aftercare, the bony defect in the anterior mandible was reconstructed with an individualized three-dimensional titanium mesh. The patient-specific titanium mesh (Yxoss CBR, ReOss, Filderstadt, Germany) was manufactured according to a 3D-computerized tomography (CT) scan (Figure 4). The treatment option, primarily an autologous bone transplantation from the iliac crest, was discussed with the patient. Due to the surgical therapy the patient already had to overcome, she refused harvesting of autologous bone from a second surgical site. Alternatively, she agreed to the suggested attempt to heal, based on a combination of a bone substitute material (BO, Bio-Oss, Geistlich Biomaterials, Wolhusen, Switzerland) and PRF prepared according to the LSCC.

A preoperatively recorded CT scan was used for manufacturing the titanium mesh according to the ideal anatomy of the mandible with respect to the nerve alveolaris inferior. Intraoperatively, blood from the cubital vein was drawn in four 10-ml glass tubes for A-PRF+ and two 10-ml plastic tubes for i-PRF (Process for PRF, Nice, France). To process the liquid i-PRF, plastic tubes were used to prevent the blood from coagulating. The solid A-PRF was processed by centrifugation of the blood in glass tubes. The tubes were placed in a centrifuge (Duo centrifuge; Process for PRF) and immediately centrifuged according to the LSCC established protocols (i-PRF: 60 g for 3 min; A-PRF+: 208 g for 8 min). The resulting A-PRF+ clots and the liquid i-PRF were isolated and combined with BO. The mesh was filled with the combination of BO, solid and fluid PRF (A-PRF+ and i-PRF; see Figures 5 through 7). In general, anesthesia was performed in a lingually positioned incision of the gingiva. After raising the mucoperiosteal flap and exposing the defect, the bone surface was slightly degloved to penetrate the cortical bone and increase bleeding from the jaw. After a trial positioning of the titanium mesh, a periosteal incision was performed to increase flap mobility that would allow wound closure after final placement of the titanium mesh. The mesh was then filled with a combination of the xenogeneic bone substitute material and PRF, prepared according to the LSCC. Afterward, the mesh was covered with a three-dimensional collagen matrix (Mucograft, Geistlich Biomaterials), which had demonstrated favorable tissue regeneration in our previous study. Finally, pressed A-PRF+ clots in the shape of membrane-like constructs were used to cover the collagen matrix above the titanium mesh. Apical matrix sutures were applied to provide a tension-free flap closure.

No complications or soft tissue dehiscences were observed during the healing process. After an integration period of 8 months, the titanium mesh was removed and 6 implants (Straumann bone level, Straumann AG, Basel, Switzerland) were placed in the augmented region (Figures 8 and 9). Concurrently with the implant placement, bone biopsies were harvested with a trephine bur for histological investigation. After another healing period of 6 months, the implants were uncovered in combination with a vestibuloplasty to increase the peri-implant attached gingiva. A split-thickness flap was prepared and moved apically, while the peri-implant mucosa was covered with a three-dimensional collagen matrix (Mucograft, Geistlich Biomaterials). During an initial healing period of 5 days, an implant-retained splint was incorporated to prevent early relapse of the mucosa. Six weeks later, the implants were uncovered, and the final removable telescopic-retained prosthetics could be incorporated (Figure 10).

The histological samples were fixed in 4% formalin, decalcified, dehydrated in alcohol, and embedded in paraffin, according to previously described methods. Afterward, the samples were cut with a microtome in sections of 4-μm thickness and stained with hematoxylin and eosin and Azan blue, as previously described.

**Discussion and Preview**

By combining the xenogeneic bone substitute material (BO), which serves as an osteoconductive scaffold, the autologous fibrin-based liquid and the solid matrices, together with an individual three-dimensional titanium mesh, could regenerate a relatively large defect in the mandible even though the defect was caused by cancer resection. Furthermore, placement of dental implants could be successfully performed and oral...
FIGURES 1–7. **FIGURE 1.** Autologous fibrin matrix platelet rich fibrin after centrifugation and subsequent compression. **FIGURES 2 AND 3.** Radiographic (Figure 2) and clinical (Figure 3) baseline situations after tumor therapy and before augmentative reconstruction. **FIGURE 4.** Three-dimensional design of the titanium mesh according to the ideal anatomy of the mandible and the position of the foramen mentale. **FIGURES 5 AND 6.** Augmentation process with the titanium mesh, a combination of the xenogeneic Bio-Oss, advanced platelet rich fibrin, and injectable platelet rich fibrin. **FIGURE 7.** Postoperative X-ray image after reconstruction of the mandibular defect.
FIGURES 8–11. FIGURE 8. Clinical situation after removal of the titanium mesh and insertion of the implants in the regenerated mandible after a mean integration phase of the augmentation (8 months). Implants could be placed with high primary stability and in full length. FIGURE 9. Postoperative X-ray image after implant insertion in the regenerated mandible. FIGURE 10. Final prosthetic rehabilitation of the regenerated mandible. FIGURE 11. Histological analysis of the extracted bone biopsies. (a) Total scan of the biopsy with new bone formation reaching all parts of the biopsy (Azan staining, total scan ×4 magnification, scale bar = 500 μm). (b) New bone formation in
rehabilitation achieved. The preoperative planning, the individually tailored titanium mesh, and the combination of solid and liquid PRF matrices results in a reduction in surgery time, postoperative pain, and a healing period compared to autologous bone transfer from the iliac crest.

The applied combined technique allowed three-dimensional bone regeneration without autologous bone transplantation from the iliac crest is the therapy of choice worldwide but can be avoided. The conventional treatment for defects is autogenous transplantation in patients with a cancer history or as a mixture of biomaterials with autogenous bone in patients without cancer history. No studies or case reports were found in the literature that used biomaterials in this specific combination protocol. Additionally, the use of titanium mesh in recent years was reported in many cases accompanied by early or late soft tissue dehiscence and mesh exposure. The meshes used were different from that in the presented case. Previously, the titanium meshes were not patient-specific but were prefabricated. Sumida et al demonstrated in a 2015 clinical trial that the use of patient-specific titanium mesh significantly reduced operation time and had a lower rate of mesh exposure compared to prefabricated meshes. However, mesh exposure resulted in the flap-preparation technique used. In 2017, Sagheb et al used patient-specific titanium mesh in combination with a mixture of biomaterials and autogenous bone to regenerate three-dimensional bone defects. Despite the application of patient-specific meshes, mesh exposure occurred in 33% of the cases.

In the present case report, we introduced a novel surgical technique and a combination protocol using a computer-aided design technique for the titanium mesh, as well as specific solid and liquid PRF protocols to regenerate three-dimensional bone defects in patients with limited regenerative potential due to a history of cancer.

For a specific patient pool that undergoes multiple surgical interventions, such as head and neck cancer patients, the alternative to autologous bone transplantation presented in this study could be an effective alternative concept in oral and maxillofacial surgery. The applied PRF matrices include regenerative components such as platelets, leukocytes, their growth factors, and fibrin as a harboring scaffold to increase the biological capacity of the bone substitute material. This combination may be the most useful for biomaterials to mainly induce a mononuclear-cell–triggered cellular tissue reaction. Recent studies demonstrated that BO induced a mild physiological tissue reaction with only minimal multinucleated giant cells, while at the same time promoting neovascularization within the implantation bed.

Numerous preclinical studies have demonstrated the capacity of PRF-matrices, especially those prepared with reduced RCF according to the LSCC to release high concentrations of different growth factors. These factors include epidermal growth factor (EGF), which is essential for vascularization and regeneration, platelet-derived growth factor (PDGF), which plays a significant role in osteogenesis, as well as vascular endothelial growth factor, which plays a major role for angiogenesis. Additionally, benefits of PRF-treated osteoblasts and fibroblasts include significantly enhanced proliferation, cell migration, and metabolic activity when compared with untreated osteoblasts and fibroblasts. Therefore, the addition of the applied PRF-based matrices (A-PRF+ and i-PRF) to BO might result in accelerated migration of osteoprogenitor cells in the augmentation bed through bioactive growth factor release, thus increasing the regenerative capacity of BO.

The histological analysis of the bone biopsies harvested at the same time as implant placement revealed good integration of the BO particles in the surrounding tissue (Figure 11a and b). New bone formation was detected to be in close relation to the BO-granules. In all analyzed biopsies, new bone formation was observed throughout the biopsy, and the histological analysis showed the increased vascularization of the connective tissue within the augmentation bed compared with the residual bone (Figure 11c). The vascularization of an implantation bed is of striking importance for tissue regeneration and new bone formation since the newly formed vessels provide nutrition and progenitor cells to the impaired region.

In conclusion, the addition of both PRF-based matrices prepared according to LSCC may act as a drug delivery system to provide the augmentation area with the required platelets, leukocytes, and growth factors to promote vascularization. Therefore, subsequent new bone formation occurs, especially in cases of large bone defects where new bone must be generated along a relatively large area.

The presented case reports the reconstruction of a severe defect in the mandible without autologous bone transfer. This presented technique might be able to replace the use of autologous bone transfer in comparable advanced defects. Further systematic analyses are necessary to determine indications for the presented technique and its limitations. Finally, we must critically examine to what extent the autologous bone blocks maintain their regenerative capacity after transplantation.

**Conclusion**

The presented case report introduces a promising treatment option for reconstruction of a severe defect in the mandible after squamous cell cancer resection and subsequent tumor aftercare. The addition of solid and fluid PRF matrices (gain by low-speed centrifugation) to a xenogeneic bone substitute material that induces a physiological and mainly mononuclear-cell–triggered tissue reaction under an individualized titanium mesh, might significantly benefit the field of tissue engineering as a potential alternative to autologous bone transfer in oral and maxillofacial surgery.

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direct contact with the BO granules at a higher magnification (Azan staining, ×200 magnification, scale bar: 100 μm). (c) granules embedded in vessel-rich connective tissue (Azan staining, ×400 magnification, scale bar = 20 μm). BO indicates Bio-Oss particles; NB, newly built bone; CT, connective tissue, black arrows, newly built bone; red arrows, newly formed vessel.
Individualized Titanium Mesh Combined With Platelet-Rich Fibrin

ABBREVIATIONS

A-PRF: advanced platelet rich fibrin
BO: Bio-Oss
CT: computerized tomography
EGF: epidermal growth factor
GBR: guided bone regeneration
i-PRF: injectable platelet rich fibrin
LSCC: low-speed centrifugation concept
PDGF: platelet-derived growth factor
PRF: platelet rich fibrin
RCF: relative centrifugation force

REFERENCES