Pre-clinical (*in vitro* & *in vivo*) studies

1. Porcine dermis-derived collagen membranes induce implantation bed vascularization via multinucleated giant cells: a physiological reaction?
   Barbeck M, Lorenz J, Grosse Holthaus M, Raetscho N, Kubesch A, Booms P, Sader R, Kirkpatrick CJ, Ghanaati S. J Oral Implantol. 2015; 41(6):e238-51.

   https://www.ncbi.nlm.nih.gov/pubmed/25546240

   In this study, the tissue reactions to 2 new porcine dermis-derived collagen membranes of different thickness were analyzed. The thicker material (mucoderm®) contained sporadically preexisting vessel skeletons and fatty islands. The thinner membrane (collprotect®) had a bilayered structure (porous and occlusive side) without any preexisting structures. These materials were implanted subcutaneously in mice to analyze the tissue reactions and potential transmembranous vascularization. Histological and histomorphometrical methodologies were performed at 4 time points (3, 10, 15, and 30 days). Both materials permitted stepwise connective tissue ingrowth into their central regions. In the mucoderm® matrix, newly built microvessels were found within the preexisting vessel and fatty island skeletons after 30 days. This vascularization was independent of the inflammation-related vascularization on both material surfaces. The collprotect® membrane underwent material disintegration by connective tissue strands in combination with vessels and multinucleated giant cells. The histomorphometric analyses revealed that the thickness of mucoderm® did not decrease significantly, while an initial significant decrease of membrane thickness in the case of collprotect® was found at day 15. The present results demonstrate that the 2 analyzed collagen membranes underwent a multinucleated giant cell-associated vascularization. Neither of the materials underwent transmembraneous vascularization. The microvessels were found within the preexisting vessel and fatty island skeletons. Additional long-term studies and clinical studies are necessary to determine how the observed foreign body giant cells affect tissue regeneration.

2. Collagen Membranes Adsorb the Transforming Growth Factor-β Receptor I Kinase-dependent Activity of Enamel Matrix Derivative.
   Ståhli A, Miron RJ, Bosshardt DD, Sculean A, Gruber R. J Periodontol. 2016; 87(5):583-90.

   https://www.ncbi.nlm.nih.gov/pubmed/26777762

   The aim of this study is to evaluate the ability of two CMs and a collagen matrix to adsorb the activity intrinsic to EMD that provokes transforming growth factor (TGF)-β signaling in oral fibroblasts.
   METHODS: Three commercially available collagen products were exposed to EMD or recombinant TGF-β1, followed by vigorous washing. Oral fibroblasts were either seeded directly onto collagen products or were incubated with the respective supernatant. Expression of TGF-β target genes interleukin (IL)-11 and proteoglycan 4 (PRG4) was evaluated by real time polymerase chain reaction. Proteomic analysis was used to study the fraction of EMD proteins binding to collagen.
   RESULTS: EMD or TGF-β1 provoked a significant increase of IL-11 and PRG4 expression of oral fibroblasts when seeded onto collagen products and when incubated with the respective supernatant. Gene expression was blocked by the TGF-β receptor I kinase inhibitor SB431542. Amelogenin bound most abundantly to gelatin-coated culture dishes. However,

incubation of palatal fibroblasts with recombinant amelogenin did not alter expression of IL-11 and PRG4.
CONCLUSION: These in vitro findings suggest that collagen products adsorb a TGF-β receptor I kinase-dependent activity of EMD and make it available for potential target cells.

3. Enhanced periodontal regeneration using collagen, stem cells or growth factors. (Bredent study)

Basan T, Welly D, Kriebel K, Scholz M, Brosemann A, Liese J, Vollmar B, Frerich B, Lang H. Front Biosci (Schol Ed). 2017;9:180-193.

https://www.ncbi.nlm.nih.gov/pubmed/27814584

The aim of the present study was to examine the regenerative potential of a) different collagen support versus blank, b) different collagen support +/- a growth factor cocktail (GF) and c) a collagen powder versus collagen powder + periodontal ligament stem cells (PDLSCs) comparatively in a large animal model. The stem cells (SC) were isolated from extracted teeth of 15 adult miniature pigs. A total of 60 class II furcation defects were treated with the materials named above. Concluding, a histological evaluation followed. A significant increase in regeneration was observed in all treatment groups. The new attachment formation reached a maximum of 77 percent. In the control group a new attachment formation of 13 percent was observed. The study shows that all implanted materials improved periodontal regeneration, though there were no significant differences between the experimental groups. Within the limitations of this study, it can be assumed that the lack of significant differences is due to the complexity of the clinical setting.

*Study refers to Angiopore (Bredent medical), which was a former private label of collprotect® membrane.

4. Use of Collagen, PTFE and PRF Membranes in Bone Reconstruction an Experimental and Histomorphometric Study.

Neculae II, Anghelescu VM, Zurac SA, Dinca OM, Vladan CG, Bucur A. J Transl Med Res. 2017; 22(1): 42-47.

https://www.sgo-iasgo.com/pdfs/2017-1-42.pdf

This study presents a comparison between outcomes of bone regeneration, after producing standardized bone defects followed by covering them with membranes, on an animal experimental model. The study was conducted on 18 New Zeeland rabbits, by creating 2 defects in the left tibial bone of each rabbit: one standardized defect with a diameter of 4 mm, and the second by creating 5 monocortical holes with a small round bur. The defects were augmented with bovine bone, beta-tricalcium phosphate and perioglass and they were covered with 3 types of membrane: collagen (12 defects - group A), PTFE membrane (12 defects - group B) and PRF membrane, made from the blood of the same rabbit (12 defects – group C). The animals were sacrificed after 6 months and analysed histomorphometrically. The new bone around graft particles has a thickness of 98.26 µm for collagen membrane, 49.19 µm for PTFE membrane and 63.98 µm for PRF membrane. The density of osteoblasts and osteocytes has an average of 0.0012 for collagen membrane, 0.0009 for PTFE membrane and 0.0010 for PRF membrane. Regarding the collagen membrane, it is observed that when used the bone. Regeneration appears to have a higher density of osteoforming cells and a higher quantity of new bone.

Clinical studies and case series

5. The concept of Screw-Guided Bone Regeneration (S-GBR). Part 2: S-GBR in the severely resorbed preimplant posterior mandible using bone xenograft and Leukocyteand Platelet-Rich Fibrin (L-PRF): a 5-year follow-up.

   Toeroek R, and Dohan Ehrenfest DM. POSEIDO. 2013; 1(2): 85-92

   http://www.poseido.info/publication/volume-1-2013/poseido-20131285-92-toeroek.pdf

   A specific form of GBR was developed using screws as space maintainers and regenerative pillars for the protection and bone growth orientation of the bone regenerative compartment, and was termed Screw-Guided Bone Regeneration (S-GBR). This approach appeared particularly adapted to the posterior mandible sites, as the screws are efficient support and protection for the bone regenerative chamber against the various mechanical constraints. This form of GBR can be associated with non-resorbable or resorbable membranes and various combinations of bone materials, but the use of Leukocyte- and Platelet-Rich Fibrin (L-PRF, Intra-Spin system, Intra-Lock, Boca-Raton, FL, USA) membranes became a very logical addition to any S-GBR protocol.

   *Study refers to BoneProtect Guide (Dentegris), which is a private label of collprotect® membrane.

6. Comparison of two different xenografts in bilateral sinus augmentation: radiographic and histologic findings.

   Panagiotou D, Özkan Karaca E, Dirikan İpçi Ş, Çakar G, Olgaç V, Yılmaz S. Quintessence Int. 2015 ; 46(7):611-9.

   https://www.ncbi.nlm.nih.gov/pubmed/25699296

   The aim of this study was to evaluate the radiographic and histomorphometric results of two different xenografts in bilateral sinus augmentation in patients with posterior maxillary atrophy.
   METHOD AND MATERIALS: Eight patients with less than 5 mm residual alveolar bone height were included in this study. One side was augmented with bovine bone graft-1 and the other side with bovine bone graft-2. Radiographic analyses were performed before and after augmentation, and before the implant placement. After 8 months of healing period, bone biopsies were obtained during implant placement.
   RESULTS: No statistically significant difference was found between the groups, based on post-augmentation and pre-implantation graft heights (P>.05). Histomorphometric evaluation demonstrated 24.63 % and 29.13 % newly formed bone in the graft-1 and graft-2 groups, respectively. Intergroup differences were not significant for the mean percentage of new bone formation (P > .05).
   CONCLUSION: Within the limitations of this study, it can be concluded that xenograft materials resulted in satisfactory bone height and trabecular new bone formation, and they could be used for the rehabilitation of atrophic maxillae.

7. Effect of sex-hormone levels, sex, body mass index and other host factors on human craniofacial bone regeneration with bioactive tricalcium phosphate grafts.

Knabe C, Mele A, Kann PH, Peleska B, Adel-Khattab D, Renz H, Reuss A, Bohner M, Stiller M. Biomaterials. 2017; 123:48-62.

https://www.ncbi.nlm.nih.gov/pubmed/28160669

The aim of this study was to elucidate the associations between these factors and bone formation after sinus floor augmentation procedures (SFA) utilizing a bioactive tricalcium phosphate (TCP) bone grafting material. We conducted a prospective study in a human population in which 60 male and 60 female participants underwent SFA and dental implant placement using a staged approach. BMI as well as levels of serum estradiol (E2), total testosterone (TT), and the free androgen index (FAI) were measured by radioimmunoassay and electrochemoluminescent-immunoassay. At implant placement, 6 months after SFA, bone biopsy specimens were harvested for hard tissue histology, the amount of bone formation was evaluated by histomorphometry and immunohistochemical analysis of osteogenic marker expression. The Wilcoxon rank-sum U test, Spearman correlations and linear regression analysis were used to explore the association between bone formation and BMI, hormonal and other host factors. BMI and log E2 were significantly positively associated with bone formation in male individuals ($p < 0.05$). Histomorphometry revealed trends toward greater bone formation and osteogenic marker expression with non-smokers compared to smokers. In male patients, higher E2 levels and higher BMI enhanced TCP stimulated craniofacial i.e. intramembranous bone repair.

8. Effect of Deproteinized Bovine Bone Mineral at Implant Dehiscence Defects Grafted by the Sandwich Bone Augmentation Technique.
   Wen SC, Fu JH, Wang HL. Int J Periodontics Restorative Dent. 2018.

   https://www.ncbi.nlm.nih.gov/pubmed/29240209

   Aim of this study was to compare the amount of radiographic horizontal buccal bone thickness (BBT) at implant dehiscence defects grafted with the sandwich bone augmentation (SBA) and modified sandwich bone augmentation (MSBA) techniques. Compared to the SBA technique, the MSBA approach involved an additional outer layer of deproteinized bovine bone mineral (DBBM) to maintain the space for bone regeneration for longer periods. A total of 19 patients, each with a buccal implant dehiscence defect, were recruited. The control group was treated with SBA technique (n = 10), while the test group was treated with MSBA technique. Cone beam computed tomography (CBCT) scans, taken at three time points (before and immediately after implant surgery, and 6 months post-treatment) were used to assess the BBT at the implant platform (-1.8 mm), the rough-smooth junction (0 mm), and 2, 4, 6, 8, and 10 mm apical to the rough-smooth junction. At 6 months post-surgery, the mean BBT in control and test groups was 1.69 ± 0.38 mm and 2.55 ± 0.21 mm, respectively. Mean BBT was significantly greater in the test group at 2, 4, 6, and 8 mm apical to the rough-smooth junction. There was no statistical difference in the mean BBT at the implant platform, the rough-smooth junction, and 10 mm apical to the rough-smooth junction between the two groups (P> .05). Within the limitations of this study, it was concluded that the additional layer of DBBM enhanced BBT along the implant, except at the smooth collar.