Augmentation of bone defect healing using a new biocomposite scaffold (PLA/TCP)

INTRODUCTION: Autologous bone graft is the current gold standard treatment for bone loss. However, disadvantages, namely limited availability and donor site morbidity, remain problematic. Several osteoconductive scaffolds based on polymers or ceramics, have been devised as bone graft substitutes. Calcium phosphate materials such as hydroxyapatite (HA) or tricalcium phosphate (TCP) have good biocompatibility as well as excellent osteoconductive properties. Despite these advantages there are some considerable drawbacks, including unsuitable mechanical properties such as brittleness, low strength and poor fatigue resistance. The incorporation of these calcium phosphate materials into a polymer scaffold could result in a composite bioactive scaffold with improved mechanical properties. Therefore, a novel poly lactate (PLA)/β-TCP scaffold was developed for bone repair (1). The developed scaffold demonstrated good in vitro biocompatible properties (2) as well as promising in vivo results in a rat study (3). The aim of this study was to demonstrate the biocompatibility and osteoconductive properties of this new PLA/β-TCP biocomposite in a large animal model over a period of 2, 4 and 12 months and to determine new bone formation, implant degradation and inflammation compared to a clinically used bone substitute and to an untreated (empty) control.

MATERIALS AND METHODS: Scaffolds: The new biocomposite is obtained by a two step process, namely melt extrusion and supercritical CO2 foaming as described previously (1). ChronOS™ (β-TCP) is a clinically used bone substitute and was used as a positive control.

Animal model and study design: Bilateral defects with a diameter of 5.1 mm and a depth of 15 mm were created within the cancellous bone of the medial femoral condyle and the proximal tibial metaphysis of 12 mature female Swiss alpine sheep (average weight: 50 kg). The three femoral and two tibial defects were filled with either the new PLA/β-TCP biocomposite, ChronOS™ or left empty. At 2, 4 and 12 months respectively, 4 sheep per group were sacrificed and samples from the femora were analysed quantitative – and qualitatively using µCT and histology. Samples from the tibia were analysed for their mechanical properties by ultrasound measurements (data not available yet).

Surgical procedure and harvest of samples: Bilateral surgical intervention was performed under general anesthesia. The correct positioning of the defects within cancellous bone was ensured by a custom made jig. A 5.1 mm drill bit with a depth regulation of 15 mm was used to create the defects. At sacrifice, implants were harvested using a coring device with an outer diameter of 12.5 mm. The resulting core consisted of the implant/defect in the centre with approximately 3 mm of surrounding cancellous bone.

Analysis: The scans were performed using high resolution settings (18 µm). The bone volume to total volume (BV/TV) was evaluated and 3D reconstructions of representative samples created. Transverse sections through the centre of the defect were stained with Toluidine blue and Giemsa-Eosin. All stained sections were evaluated qualitatively for new bone formation, implant degradation as well as signs of inflammation and/or immunologic reaction. For quantitative analysis microradiographs of the sections were digitized. Relative bone formation was assessed as bone density (BD) using a custom made macro.

Statistical analysis: Statistical analysis was carried out on the histomorphometrical data. Repeated measures ANOVA was performed for every variable with time as between-subject factor and treatment as within-subject factor. Bonferroni Post-hoc tests were performed to compare between treatment groups. In contrast to histomorphometry, CT data from both sides could not be treated independently so that only descriptive statistics was used (n=4).

RESULTS: μCT: At 12 months, BV/TV of defects treated with the biocomposite was 22.1%, of the untreated (empty) control was 16.7%, and for ChronOS™ 45.4%. On the 3-Dimensional reconstructions bone formation was seen throughout the biocomposite, whereas bone formation within the untreated control was restricted to the border of the defect (figure 1).

DISCUSSION: In this study, the sheep as animal model was chosen since loading, bone structure and bone remodelling are similar to that of humans. At 2 and 4 months a mild, locally restricted foreign body reaction was detected around the biocomposite and integration of new formed bone was hindered by a fibrous layer surrounding the implant. However, at 12 months the biocomposite was well integrated and the inflammation could no longer be detected. This early non specific mild foreign body reaction to PLA and its consecutive resolution has been described (4).

In comparison to the untreated defects the overall BD within the defect for the biocomposite and the empty control were similar, but the distribution of the newly formed bone was different. At 12 months new bone with a structure similar to the surrounding cancellous bone had formed throughout the defect in samples filled with the biocomposite whereas bone formation without scaffold was restricted to the border of the defect with a dense structure. Hence, bone healing pattern within the biocomposite is qualitatively favourable compared to “untreated” healing. However, bone formation is slower compared to a TCP only scaffold. Nevertheless, this new biocomposite could be useful in clinical situations where slow bone remodelling is desirable or high strains are expected.

REFERENCES: