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Long-term *in vivo* properties of natural mineral fiber reinforced composite products

Arctic Biomaterials Research and Development

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Introduction

Many beneficial properties of silica-based natural mineral fibers are well known and well reported in the literature [1, 2]. Composite materials combine the good properties of bioactive natural mineral fibers and bioabsorbable polymers offering greatly improved mechanical properties compared to implants made of plain polymers. Although not many *in vivo* studies have been reported, some promising results have been seen [3, 4]. This white paper summarizes a two-year rabbit study of injection molded composite samples.

Materials and methods

The samples were injection molded composite pins with diameter of 2 mm and length 6 mm (Figure 1). The material was a blend of Evolvecomp GF40PLD96 and 70L/30D,L PLA (RESOMER LR 708). Natural mineral fiber content of manufactured composites was 20 wt-% and the fibers had a mean diameter of 13 µm. Samples were EtO sterilized before implantation.

Local tissue response of the samples was studied in a two-year study implanted in the lateral condyle bone of rabbits at NAMSA Northwood, Ohio, U.S.A. The time points of the study were 4, 13, 26, 52 and 104 weeks. HDPE implants were used as a non-degradable controls. All implanted sites were evaluated histologically, microscopically as well as using µCT. In addition, hematology and clinical chemistry parameters were followed. An additional µCT study was done at Tampere University.

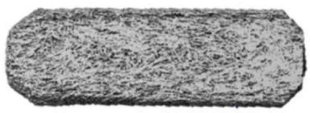

Figure 1. *µCT image of the sample.*

Results

Overall, no adverse effects of the implanted samples were observed during the two-year study. There were no biologically significant macroscopic or microscopic adverse findings regarding biocompatibility. In addition, no significant adverse findings were made in the hematologic or clinical chemistry analysis. Sample material showed similar tissue responses with the control samples at all studied time points. These responses were aligned with the expectations for implanted materials in bone and they demonstrate that the composite material shows good biocompatibility when implanted in bone tissue.

All of the implant sites were surrounded by a narrow band of fibrosis. Histological HE stains of the composite implants are shown in Figure 2 and of the controls in Figure 3. In both figures, time points of 4, 26, 52 and 104 weeks are shown. Purple areas in the figures show new bone and progressive new bone growth was seen around the composite implants. The HDPE implants that served as controls were unfortunately of different shape than the composite samples and the fit of them to the drilled bone defects was not as tight as of the composite implants. Thus the comparison of the new bone formation around the implants is not very straight forward.

µCT images of the samples at 52 and 104 week time points are shown in Figure 4 where white areas represent bone tissue. At both time points, bone growth is seen on the surface of the samples as a thin layer of bone tissue. Findings from Figure 2 support this observation (purple layers of new bone around the samples). At 52 week time point, no bone ingrowth to the implants was evident but at 104 weeks a clear bone ingrowth was observed, extending even through the whole implant (Figure 4).

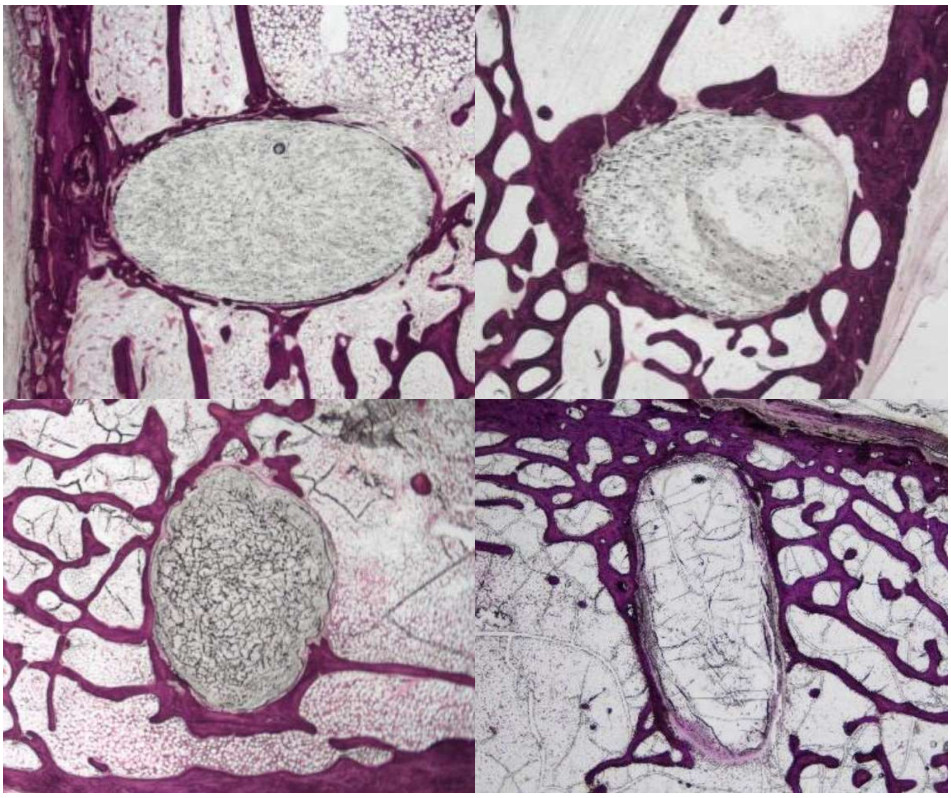

Figure 2. *HE stained overview of the implanted sample and surrounding tissues at 4 weeks (top left), 26 weeks (top right), 52 weeks (lower left) and 104weeks (lower right). Purple areas show new bone.*

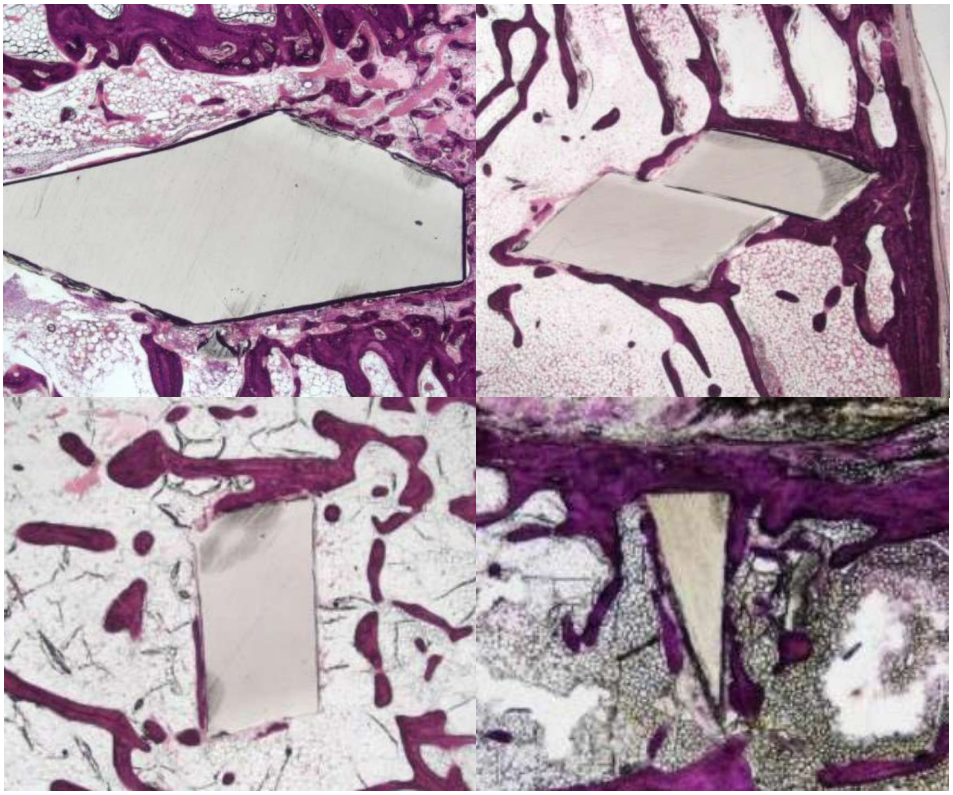

Figure 3. *HE stained overview of the implanted control sample and surrounding tissues at 4 weeks (top left), 26 weeks (top right), 52 weeks (lower left) and 104weeks (lower right). Purple areas show new bone.*

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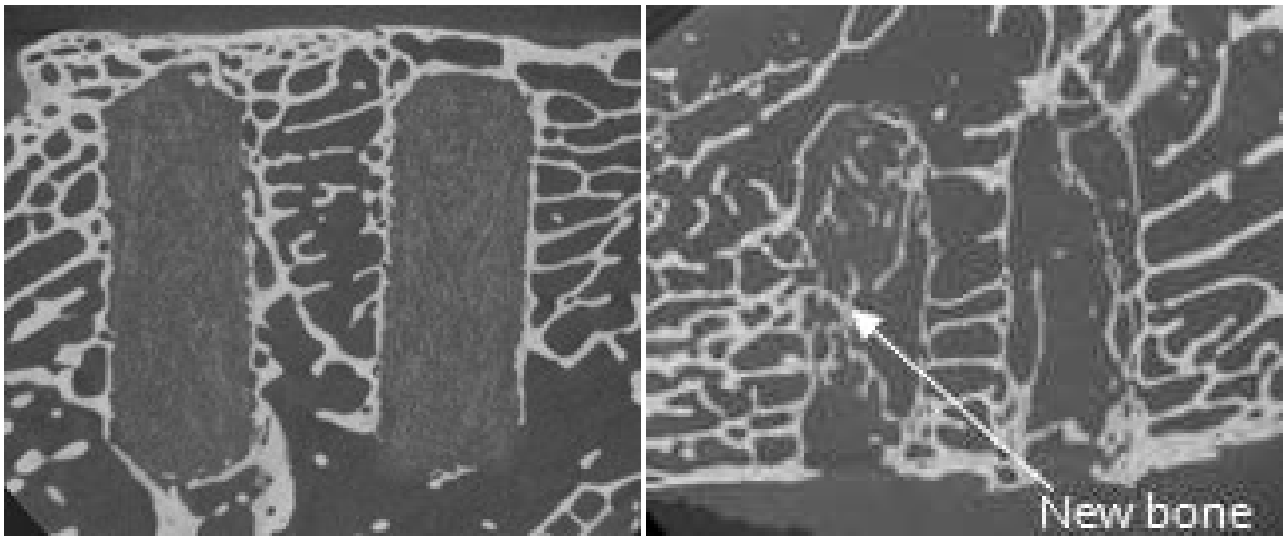

Figure 4. *µCT images of the implant at 52 (left) and 104 weeks (right). White areas represent bone tissue.*

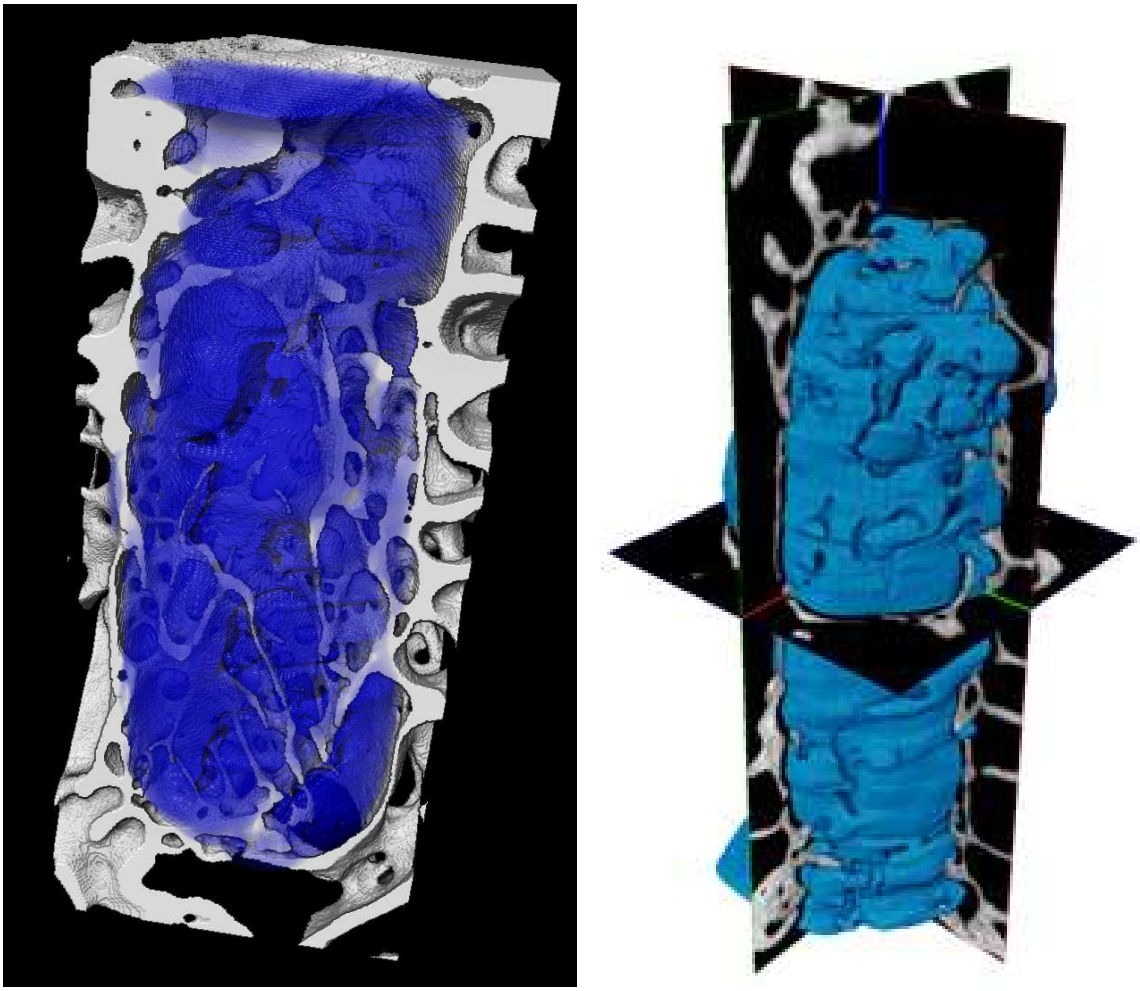

Figure 5. *Two 3D reconstructions of the µCT images at 104 week time point. Blue color represents the sample material and white color represents bone tissue. Images provided by Tampere University.*

Figure 5 shows two 3D reconstructions of the µCT imaging at 104 week time point. Implant material is represented with blue color and bone tissue is represented with white color. Bone ingrowth through the sample can be seen in the left side image and implant material degradation can be seen as grooves and dents in the implant material in the right side image.

When bioabsorbable polymers like polylactides degrade, their molecular weight decreases first as the polymer chains are cut to smaller fractions due to random scission caused by hydrolysis. In *in vitro* degradation studies, this is seen as decrease in the inherent viscosity and in the molecular weight of the polymer. When the polymer molecules are cut to small enough fractions, they start to dissolve to the surrounding fluid, which is seen as mass loss of the implant. In this study, no implants were retrieved so that the molecular weight of the polymer could be measured, thus the extent of the polymer degradation cannot be estimated. However, it has been reported in literature that the *in vivo* degradation of lactide based polymers can be estimated with *in vitro* results [5].

No complete degradation (mass loss) was observed in the implants in the time frame of this study. However, the µCT study revealed quite advanced implant degradation at 104 week time point. Some mass loss of the composite implants was seen at 104 week time point in the 3D reconstructions of the µCT images (Figure 5) as grooves on the implant surface. Additionally, the µCT images showing bone ingrowth through the implant material (Figure 4, Figure 5) suggest an exceptionally good potential for fast bone regeneration into the implant space after complete degradation of the implant material.

Similar composite materials have also been studied *in vivo* by Wu et al. [3] In their study, natural mineral fibers in the composite seem to induce favorable osteogenic action and reduced inflammatory reaction especially in the early phase after implantation. This is often considered as critical phase in the healing process. Their findings also indicated increased new bone formation and thus better anchoring of the implant to the bone in early phases of the healing period after implantation.

Conclusions

The biocompatibility and bone-growth promoting properties of the natural mineral fiber reinforced PLDLA composites were studied in a two-year rabbit study. The findings show that the composite implants do not elicit any adverse reactions in the surrounding tissues thus showing good biocompatibility. Additionally, bone ingrowth into the implants was seen although the implanted materials were not completely degraded within the time frame of this study.

This study showed that natural mineral fiber reinforced composite materials are very promising bioabsorbable materials for clinical use in bone-related applications. The results indicate good biocompatibility. However, it has to be kept in mind that the final verification of the biocompatibility and efficacy must always be performed for the final product.

References

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