Osteogenic differentiation of human bone marrow stromal cells in porous scaffolds from mineralized collagen

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INTRODUCTION: Scaffold materials with properties close to natural bone, which consists mainly of collagen type I and nanocrystalline hydroxyapatite are highly advantageous in bone engineering. We developed sponge-like porous scaffolds from biomimetically mineralised collagen. A first *in vivo* study revealed good biocompatibility and faster resorption compared to other HA-collagen composites [1]. In the present study we investigated the osteogenic differentiation of human bone marrow stromal cells (hBMSC) on 3D scaffolds of mineralised collagen.

METHODS: Porous 3D structures from mineralised collagen were fabricated applying a procedure in which collagen fibril reassembly and precipitation of nanocrystalline hydroxyapatite (HA) occur simultaneously [2]. hBMSC were seeded onto the 3D scaffolds and cultivated for 4 weeks in the presence and absence of osteogenic supplements. We studied viability, proliferation and osteogenic differentiation in terms of total lactate dehydrogenase (LDH) activity, DNA content and alkaline phosphatase (ALP) activity. Furthermore, the expression for bone-related genes (ALP, osteopontin, osteocalcin) was analysed by real-time PCR.

RESULTS: hBMSC and osteogenically induced hBMSC showed good proliferation rates within the three dimensional scaffolds. Determination of DNA content revealed a 2.5 to 5-fold increase of cell number after 28 d of cultivation. In coincidence to these findings we furthermore detected an increase of total LDH activity over 28 d which implies a raise of viable cells within the cultivation period.
The specific ALP activity of osteogenically induced hBMSC increased during the cultivation time of 28 d, whereas the specific ALP activity of the non induced hBMSC was not raised during the whole cultivation time.
Applying real-time PCR we detected an increase of ALP expression as well as of osteopontin expression for osteogenic induced hBMSC, but no increase of osteocalcin expression.

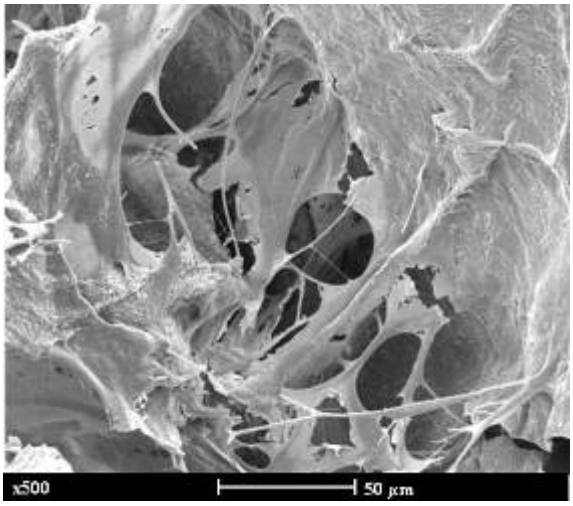

Fig. 1: SEM image of hBMSC in a porous scaffolds from mineralised collagen, 2 d after seeding

DISCUSSION & CONCLUSIONS: We conclude from our results, that the examined scaffolds from biomimetically mineralised collagen are appropriate to support proliferation as well as osteogenic differentiation of hBMSC. Due to its similarity to extracellular matrix of bone tissue this biomaterial is a promising matrix for bone tissue engineering applications

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