

ENGINEERING OF HYPERTROPHIC CARTILAGE WITH AND WITHOUT POLY(GLYCOLIC ACID) SCAFFOLDS

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INTRODUCTION: An alternative approach to bone tissue engineering could be using a tissue-engineered template that can mineralize in vitro or in vivo (e.g. hypertrophic cartilage). Cartilage has the advantage that it can survive in a relatively hypoxic environment, which may allow more time for vascularisation of the engineered graft to develop post-implantation. Development of an appropriate vasculature is essential for the maintenance and development of bone tissue. For maxillofacial reconstruction, nasal chondrocytes offer a potentially useful source of cells if they can be modulated to form hypertrophic cartilage. The aim of this research was to investigate different cell sources and conditions that promote generation of a cartilage construct with characteristics of hypertrophic tissue.

METHODS: Chondrocytes were isolated from rat sternal and nasal cartilages and their numbers expanded in monolayer culture. However, it is well known that chondrocytes lose their chondrogenic phenotypes under 2D culture conditions. Hence, when sufficient cell numbers were obtained, chondrocytes were cultured as pellets¹ or on PGA scaffolds under conditions that promote chondrogenic and osteogenic differentiation. In this 3D environment, chondrocyte re-differentiation is induced. The stage of chondrocyte differentiation was evaluated by analysis of the extracellular matrix by immunolocalisation of collagens (types I, II and X) and histochemical detection of proteoglycans, calcium deposition and alkaline phosphatase.

RESULTS: Preliminary data showed successful process of chondrogenic re-differentiation in pellets and PGA scaffolds of both cell types. High alkaline phosphatase activity and collagen X expression was detected in PGA constructs (Fig.1) and suggested that chondrocytes were entering the hypertrophic stage. No collagen type X was found in pellet cultures. Ongoing work is directed at investigating culture conditions to accelerate the process of hypertrophic differentiation in

nasal chondrocytes. Consequently research is also directed to detect other markers of terminally differentiated chondrocytes (e.g. MMP-13).

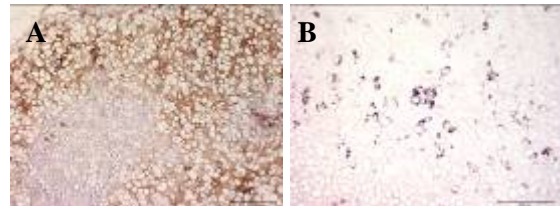


Fig. 1: Collagen type X (A) and alkaline phosphatase activity (B) in nasal chondrocytes cultured on PGA scaffolds.

DISCUSSION & CONCLUSIONS: Both nasal and sternal chondrocytes successfully re-differentiated in both culture systems. However, chondrocyte hypertrophy indicated by collagen X expression, was only observed when the cells were cultured on PGA scaffolds. This suggests that PGA scaffold could be a useful material for the generation of hypertrophic cartilage from nasal chondrocytes. That would be of a therapeutic value for maxillofacial reconstruction.

REFERENCES: ¹Y. Kato, et al (1988) "Terminal differentiation and calcification in rabbit chondrocyte cultures grown in centrifuge tubes: regulation by transforming growth factor beta and serum factors." Proc Natl Acad Sci U S A

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