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PREPARATION AND *IN VITRO* EVALUATION OF A SCAFFOLD COMPOSED OF ELASTIN AND TYPE I COLLAGEN

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INTRODUCTION: Tissue engineering is a new field of research, which aims at regenerating tissues and organs. Tissues are basically made up of cells and their extracellular matrix, and consist primarily of proteins. Cells taken out of their context will lose their shape and function. A major goal of tissue engineering is the preparation of a suitable scaffold for cells to proliferate, migrate and differentiate. The scaffold should induce cells to form the desired tissue.

Type I collagen is an extracellular matrix protein that is widely used for this purpose. It provides adhesive properties and tensile strength. Unlike type I collagen, insoluble elastin is hardly used. Elastin provides elasticity to tissues/organs and is crucial for e.g. blood vessels in order to cope with blood pressure. In this paper, we will describe the production and *in vitro* evaluation of a composite elastin-collagen scaffold.

METHODS Type I collagen was purified using various neutral salt and dilute acid extractions¹. Insoluble elastin was isolated by several extraction steps and enzyme digestions, and was devoid of microfibrillar components (in contrast to traditionally prepared elastin)². Purity is of great importance, since contaminations may lead to immunological responses.

We prepared a porous scaffold of elastin and type I collagen by controlled freezing and lyophilizing. Pores will allow cells to grow into the scaffold.

The cellular reaction to the composite matrix was evaluated *in vitro*. Human myoblasts and human lung fibroblasts were seeded on the composite scaffold, and the proliferation and differentiation were studied by light microscopy and by analysis of the mitochondrial dehydrogenase activity. In addition, specimens were studied by scanning electron microscopy (SEM).

RESULTS: Composite matrices of insoluble elastin and type I collagen were prepared by lyophilization of a diluted acid dispersion. This resulted in a porous structure with pores of about $20 \ \mu m$ (Fig. 1).



Fig. 1: SEM image of the composite porous matrix showing the interactions between elastin (large fibers) and collagen (small fibrils).

Human lung fibroblasts and human skeletal muscle cells showed similar proliferation on the composite scaffold compared to polystyrene culture dishes. Myoblasts adhere, align, and fuse to form myotubes (Fig. 2).



Fig. 2: SEM image of human myotubes on the composite porous scaffold. Cells were cultured for 14 days.

CONCLUSIONS & DISCUSSION: In this study, a composite, non-toxic, elastin-collagen scaffold was prepared, which sustained proliferation and differentiation of human cells (myoblasts, fibroblasts). Future experiments will include *in vivo* evaluation of the composite scaffold.

REFERENCES: ¹J.S. Pieper et al. (1999) *Biomaterials* **20**:847-858. ²W.F. Daamen et al. Biomaterials *in press*.

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