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Co-Culture-System of hCBEC and cardiomyocytes; first steps towards a vascularized artificial 3-dimensional cardiac patch

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Objectives:

Tissue Engineering of bioartificial cardiac tissue represents a promising method for repair of heart tissue. The need for oxygen and nutrition supply may be addressed by in vitro vascularization. This study concentrates on a cardiac construct based on a co-culture of neonatal rat cardiomyocytes (nRCm) and human cord blood derived endothelial cells (hCBEC) on biological vascularized matrix (BioVaM).

Method:

By testing several media in respect to their impact on cell morphology, proliferation and differentiation for both cell types a medium was established for co-culturing hCBECs and nRCm. Differentiation of cells was evaluated by expression of specific markers (RT-PCR). Dezellularized segments of porcine small intestine with preserved pedicles were cut open along longitudinal axis and resulting patches were sutured to silicone cushions. Vessel structures were reseeded with 5×10^7 TAMRA labeled hCBECs and submucosa with 1×10^5 nRCm/cm² one day later. After 4d cultivation in a perfusion bioreactor, constructs were examined by fluorescence microscopy to evaluate the reseeded progress.

Results:

RPMI supplemented with 10% FCS and EGM-2 Single-Quots (Lonza) showed best results for both cell types. Proliferation rate was comparable to each specific media. Cell morphology stayed well defined and the state of differentiation was preserved. Fluorescence microscopy revealed nRCm layers parallel aligned to the longitudinal axis on submucosa, as well as partially reseeded vessel structures underneath.

Conclusions:

Our construct may represent a first step towards the generation of a complex artificial vascularized cardiac construct for the therapy of injured myocardium.