Karolinska Institutet

Institutionen för molekylär medicin och kirurgi, Karolinska Institutet

# Stem cells for cardiac regeneration

AKADEMISK AVHANDLING
som för avläggande av medicine doktorsexamen vid Karolinska Institutet offentligen försvaras i föreläsningssalen C1:87, Karolinska Universitetssjukhuset Huddinge

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av

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# ABSTRACT

Recent studies have demonstrated a turnover of cardiomyocytes throughout the adult life. Whether this regenerative potential is due to proliferation and differentiation of resident stem cells or to the dedifferentiation of adult cardiomyocytes remains unclear. Generation of new cardiomyocytes is critical for cardiac repair following myocardial injury; however the mechanisms by which injury responses modulate differentiation and proliferation of cardiomyocyte progenitors is not clear.

In Study I, we show that sublethal apoptotic stimuli modulate the differentiation of mouse embryonic stem cells (ESC) into cardiomyocytes through a caspase-dependent mechanism. This indicates a direct link between caspase activation and initiation of the cardiogenic programs. Further unfolding of these mechanisms may have significant implications for the understanding of the dynamics of cardiac regeneration after myocardial injury involving both exogenous and endogenous cell sources.

The intrinsic regenerative capacity of human fetal cardiac mesenchymal stromal cells (MSCs) has not been fully characterized. In Study II our aim was to establish a culture protocol for large-scale expansion of human fetal cardiac MSCs with characteristics of cardiovascular progenitor cells. By culturing the cardiac MSCs on defined laminin (LN)-based substrata in combination with stimulation of the canonical Wnt/β-catenin pathway we could generate multi-potent cells which could differentiate into endothelial, smooth muscle cells, and spontaneously beating cardiomyocytes. The expanded cardiac MSCs stained positive for the progenitor markers: Pdgfr-α, Isl1, and Nkx2.5, and subpopulations also expressed: Tbx18, Kdr, c-Kit, and Ssea-1. Our protocol for large-scale culture of human fetal cardiac MSCs enables future exploration of the regenerative functions of these cells in the context of myocardial injury *in vitro* and *in vivo*.

Subsequently, in Study III we performed bioenergetic and metabolic profiling of human fetal cardiac MSCs as a tool for investigating metabolic changes in embryonic hearts during development. The critical role of metabolism in the active control of cell renewal and lineage specification has recently come into focus, and Wnt has been proposed as one of the signaling pathways which links energy metabolism to cell fate decision. In the MSCs derived from hearts of different gestational age, we observed an increase in oxidative metabolism during first trimester of development. Finally, stimulation of the canonical Wnt/β-catenin signaling pathway under hypoxia appeared to enhance long-term MSC expansion, and resulted in a cell population with higher cardiogenic differentiation potential. Our findings suggest that manipulation of metabolic signals may facilitate endogenous stem cell activation, tissue renewal and induction of cytoprotective properties.

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