Optimized Differentiation of Mesenchymal Stromal Cells for Vascular Tissue Engineering Applications

Corina Vater\textsuperscript{1,2}, Stefan Zwingenberger\textsuperscript{1,2}, Matthias Schieker\textsuperscript{3}, Klaus-Peter Günther\textsuperscript{1,2}, Maik Stiehler\textsuperscript{1,2}

Corresponding Author: maik.stiehler@uniklinikum-dresden.de

\textsuperscript{1}Department of Orthopedics, University Hospital Carl Gustav Carus, Dresden, Germany, \textsuperscript{2}DFG Center for Regenerative Therapies Dresden (CRTD), Germany and \textsuperscript{3}Experimental Surgery and Regenerative Medicine, Department of Surgery, LMU, Munich, Germany

Introduction

Bone marrow-derived mesenchymal stromal cells (MSCs) can differentiate into various cellular phenotypes and are key components for cell-based musculoskeletal regeneration \[1\]. Due to the limited availability and replicative capacity of somatic smooth muscle cells (SMCs), MSCs represent an appealing source of smooth muscle progenitor cells for vascular engineering approaches \[2\]. Therefore, the aim of this study was to evaluate enhancement of MSC differentiation into SMCs by different types of cell culture media.

Materials and Methods

Human immortalized single-cell derived MSCs \[3\] and primary porcine MSCs were cultured in α-MEM containing different concentrations of FCS with and without TGF-β1 for up to 14 days. DNA content was determined to assess cellular proliferation. To evaluate differentiation of MSCs into SMCs gene expression levels of α-SMA, Calponin and SMMHC were analysed by quantitative real-time RT-PCR and normalized to GAPDH expression. Furthermore cell morphology was evaluated qualitatively by light and fluorescence microscopy. Overall statistical significance was defined as \(p < 0.05\) (two-sided) based on all pairwise comparisons using one-way analysis of variance (ANOVA).

Results

Most favourable cell proliferation rates were observed for α-MEM with 10\% FCS and without TGF-β1 (increase in cell number day 1 to day 14, x-fold, hMSCs: 36.9 ± 3.4, pMSCs: 6.0 ± 2.2). In contrast, cultivation of MSCs in medium containing 0.5\% FCS and TGF-β1 lead to significantly lower cellularity observed (increase in cell number day 1 to day 14, x-fold, hMSCs: 6.8 ± 0.9, pMSCs: 1.4 ± 0.4, \(p < 0.05\)). Highest gene expression levels for α-SMA and Calponin were observed by combinations of 5\% and 0.5\% FCS, respectively, and TGF-β1.

Discussion and Conclusions

Differences in combinations and concentrations of cell culture supplements notably influence muscular differentiation of MSCs \textit{in vitro}. Our results indicate that low concentrations of FCS and the presence of TGF-β1 enhance differentiation of MSCs towards the SMC phenotype. These findings will help to support further optimization of vascular engineering strategies for cell-based musculoskeletal tissue regeneration.

References


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