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Identify hemodynamic factors implicated in differentiation of stem cells into endothelial cells Identify hemodynamic factors implicated in differentiation of stem cells into endothelial cells

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Introduction: The effects of physical environmental forces on tissue genesis and cell differentiation are largely unknown. Microfluidic devices have been used for the study of the effects of physical epigenetic factors on cell phenotype. These studies showed that fluid forces similar to blood flow may influence the differentiation of stem/progenitor cells into endothelial cells. However, the microfluidic systems that more closely resemble the hemodynamics of in vivo microvascular environment are necessary for more accurate in vitro representation of stem cell differentiation into endothelial cells. Recently, we have developed a synthetic microvascular network (SMN) on a polydothalsiloxane (PDMS) chip which accurately represents the size and vascular geometry of in vivo microvascular networks, and can mimic the flow conditions in capillary networks. Therefore SMN offers the advantage of identification of local hemodynamic parameters that are significant for vasculogenesis. In this study, by using these SMNs, we have successfully identified the hemodynamic factors implicated in differentiation of adult bone marrow–derived mesenchymal stem cells into endothelial cells.

Methodology: Undifferentiated MSCs were seeded to form the lining of SMN and subsequently were exposed to physiologically relevant flow rates (0.1, 0.2, 0.3, 0.6 μl/min) in the presence and absence of VEGF for studying the in situ differentiation of MSCs into endothelial cells. The effect on differentiation were established by a) examining the relative endothelial marker expressed in cells harvested from all experimental cultures and comparing to non-treated (static) control cells, and b) analyzing the percentage of differentiated cells with morphological characteristics of endothelial cells differentiated in each of the experimental groups and comparing to non-treated control cells. Briefly, the morphological characteristics were quantified by a Nikon Eclipse TE2000 inverted fluorescent microscope. The cells from the SMNs at different time points were harvested and immunocytochemical analysis were performed for the evaluation of dose and time- depended effects of hemodynamic factors on differentiation of MSCs into endothelial cells.

Results & Discussion: The SMNs were successfully seeded in the SMNs and MSC growth in the SMN towards confluence between 2 to 3 days after seeding. The viability assay shows more than 90% cells are viable. The MSCs cultured under different flow rates and durations, the flow rate 0.6 μl/min at 48 - 72 hrs after cell seeding induces more differentiation than other conditions. Addition of VEGF in the culture medium further increases the differentiation of MSCs into ECs. It is concluded that the optimal culture conditions for differentiation of MSCs into ECs is culturing MSCs in SMNs with VEGF at the flow rate of 0.6 μl/min for 48-72 hrs post cell seeding. The differentiation under different geometric condition (e.g. bifurcation vs. straight) was studied and the percentage of the MSCs differentiated into ECs in the straight channels is much higher than that in bifurcation. Immunocytochemical analysis further confirmed the differentiation of MSCs into ECs.

Conclusions: We have successfully identified the hemodynamic factors that can optimize the culture conditions in MSC differentiation into endothelial cells. The MSCs-derived endothelial cells exhibit the characteristics of endothelial cells found in vivo.

Biography

Bin Wang has actively pursued extramural research supports, has submitted many grant proposals and received six extramural research grants with several proposals pending. Throughout his career, he has participated in 52 national and international conferences. He has also authored three book chapters in Bionic Human and published 18 peer-reviewed papers in reputable journals. He is an active peer reviewer for both grant review panels and scientific journals, having reviewed more than 150 grant proposals.

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