

used to replace those cells, in which induced pluripotent stem cells (iPSCs) have the great potential. iPSCs are an attractive option to derive new, fully functional ECs to regenerate damaged vasculature.

**Methods** In this study we have generated iPSCs from human fibroblasts using viral induction of the four reprogramming transcription factors (OCT4, SOX2, KLF4, and c-MYC) or using a DNA-free integration method based on a single plasmid transfection and neomycin selection. iPSCs generated by plasmid transduction and selection was faster and provided a higher efficiency of colony formation. Colonies obtained using both methods were isolated, expanded and fully evaluated according to the standard criteria of iPSCs characterisation, including morphology, pluripotent marker expression, alkaline phosphatase staining, and teratoma formation.

**Results** Human iPSCs were differentiated towards vascular cell lineages and specific to ECs. To this end, human iPSCs were seeded on collagen IV and cultured in the presence of Vascular Endothelial Growth Factor (VEGF). Time course experiments revealed the ability of iPSCs to differentiate into ECs and also revealed a greater potential to derive ECs from pluripotent cells generated by transient transfection. In an attempt to obtain a pure population of ECs, iPSCs cells were pre-differentiated into an early progenitor stage and CD34 selection was performed. The CD34<sup>+</sup> cells were differentiated into ECs demonstrating a higher efficiency and led to greater homogeneity in derived ECs, expressing in high levels endothelial markers such as VE-Cadherin, PECAM1, KDR, eNOS, vWF. During iPSCs differentiation into ECs, the gene SETSIP (similar to SET protein) was found to be up-regulated. In order to define the mechanistic role of SETSIP, the protein was over-expressed and this markedly enhanced EC differentiation. Conversely, when SETSIP was knockdown by shRNA, EC differentiation was significantly suppressed. Importantly, immunofluorescence staining showed a nuclear translocation for SETSIP during EC differentiation and subsequent luciferase assays indicated that SETSIP and not SET protein induced VE-Cadherin transcriptional activation in the ECs.

**Conclusion** In this study we have shown that CD34 positive cells derived from pre-differentiated human iPSCs have the potential to differentiate into ECs. SETSIP appears to have a critical role in this process though control of VE-Cadherin transcriptional activation. Ongoing studies that identify the key mechanisms controlling iPSC-derived EC-differentiation have considerable relevance for regeneration of damaged vasculature in a range of diseases.

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**HUMAN INDUCED PLURIPOTENT STEM CELLS DIFFERENTIATE INTO PURE POPULATIONS OF ENDOTHELIAL CELLS MEDIATED BY SET SIMILAR PROTEIN THROUGH VE-CADHERIN TRANSCRIPTIONAL ACTIVATION**

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**Brief Introduction** Cardiovascular disease is a leading cause of death, while the progression of the disease is closely related to the damaged endothelial cells (ECs) on the vessel wall. Regenerative medicine is now focused on the generation of functional cells to be