Supplementary information for

**High-Purity Production of Endothelial Cells from Human Pluripotent Stem Cells**

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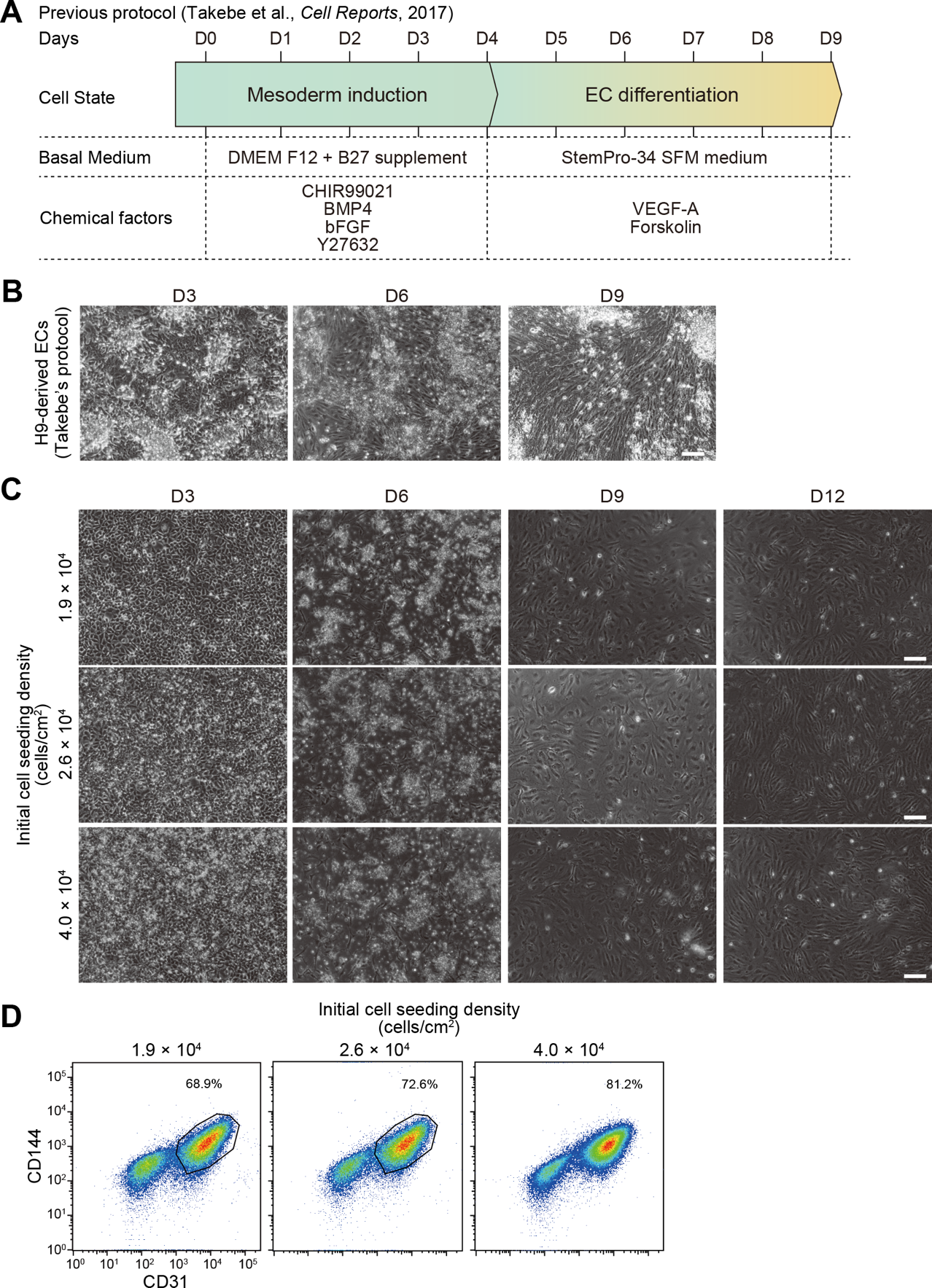
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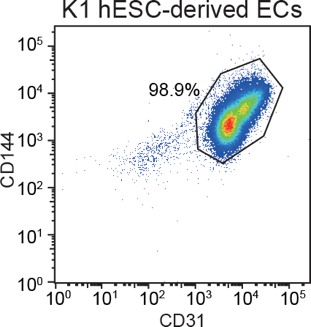
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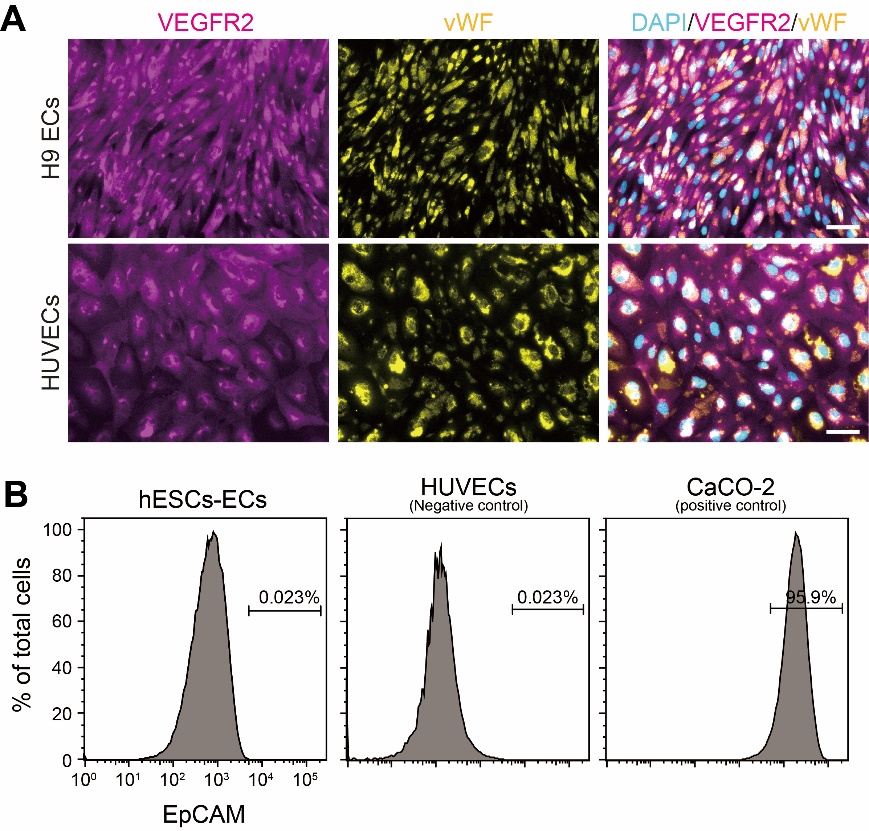
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**Supplementary Figure S1:** Comparative analysis for quality of endothelial cells (ECs) derived from H9 hESCs, by using the protocols in this study and the one developed by Takebe et al., *Cell Reports*, 2017. endothelial differentiation protocols. (**A**) A schematic illustration of the Takebe’s protocol to differentiate hESCs to ECs. (**B**) Representative micrographs of H9 hESC-derived ECs obtained with Takebe’s protocol. A scale bar represents 100 µm. (**C**) Representative micrographs of H9 hESC-derived ECs by using our protocol with three initial cell seeding densities. A scale bars represent 100 µm. (D) Flow-cytometric analysis for VE-cadherin (CD144) and PECAM-1 (CD31) in ECs with three initial cell seeding densities. Percentiles of CD144+ CD31+ cells are noted for each graph.

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**Supplementary Figure S2:** Flow-cytometric analysis for CD144 (VE-cadherin) and CD31 (PECAM-1) in K1 hESCs-derived ECs.



**Supplementary Figure S3:** **Additional characterization of endothelial and epithelial markers. (A)** Immunofluorescence staining of hESC-derived endothelial cells (ECs) for the markers VEGFR2 (magenta) and von Willebrand Factor (vWF) (yellow). Nuclei are counterstained with DAPI (cyan). Scale bar represents 100 µm. (**B**) Representative flow cytometry histogram illustrating the surface expression of the epithelial marker EpCAM on H9 hESC-derived ECs. HUVECs and CaCO-2 cells were used as negative and positive controls of the epithelial marker EpCAM, respectively.

**Supplementary Table S1:** Gene list of K-means clustered most variable genes from BRB-seq transcriptional comparison of hESC-derived endothelial cells, hESCs, and HUVECs revealed four gene sets with distinct pathway enrichments

**Supplementary Table S2:** Enriched Gene Ontology terms with K-means clustered most variable genes from BRB-seq transcriptional comparison of hESC-derived endothelial cells, hESCs, and HUVECs revealed four gene sets with distinct pathway enrichments

**Supplementary Table S3:** Enriched pathways with K-means clustered most variable genes from BRB-seq transcriptional comparison of hESC-derived endothelial cells, hESCs, and HUVECs revealed four gene sets with distinct pathway enrichments