

Figure S1

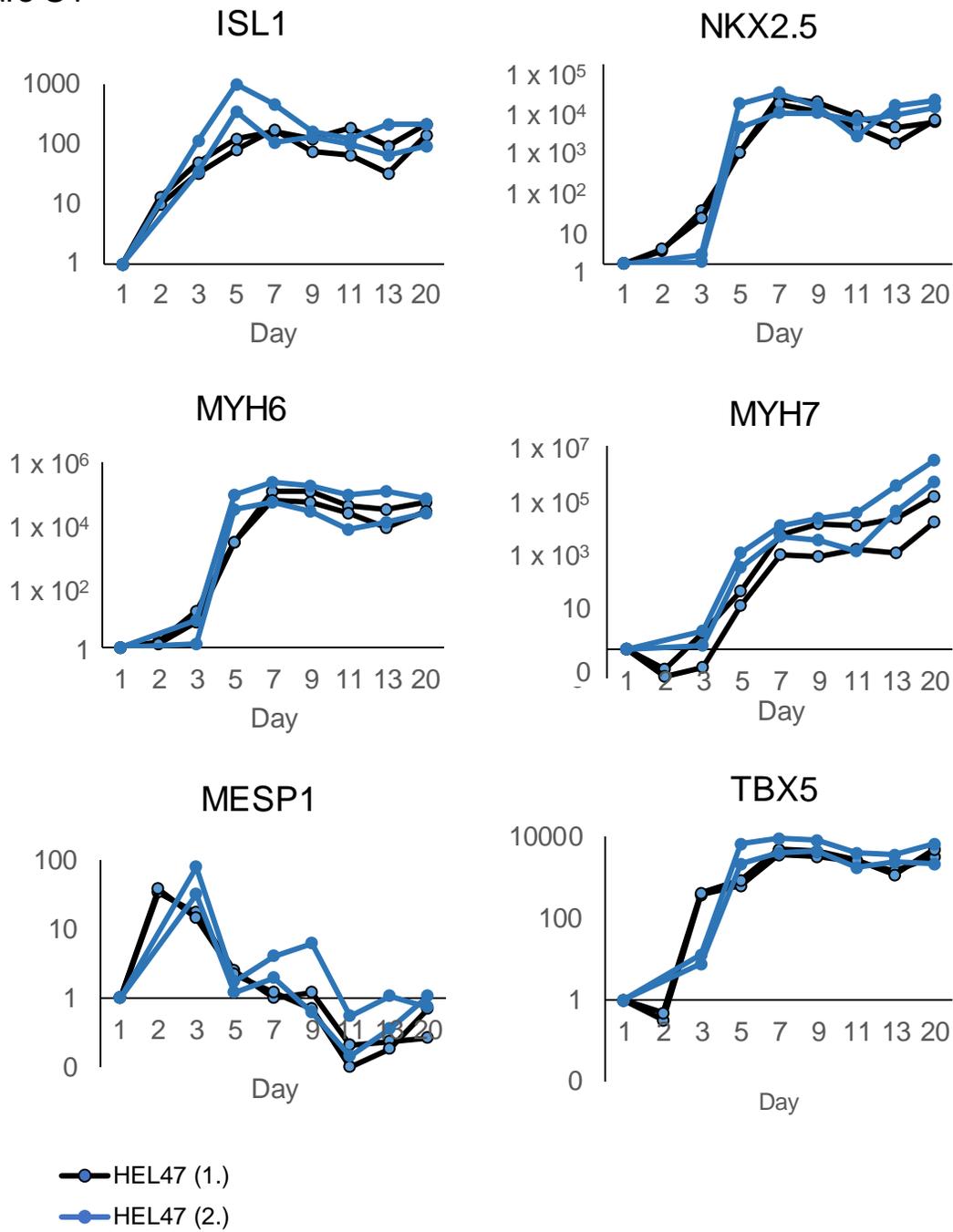


Figure S1: Expression of cardiomyocyte markers during differentiation. qPCR analysis shows that CM marker gene expression is increased during differentiation. Two differentiation experiments (HEL47.2 (1.) and HEL47.2 (2.)) with two replicates each are shown. Expression levels have been normalized by comparing to day 1.

Figure S2

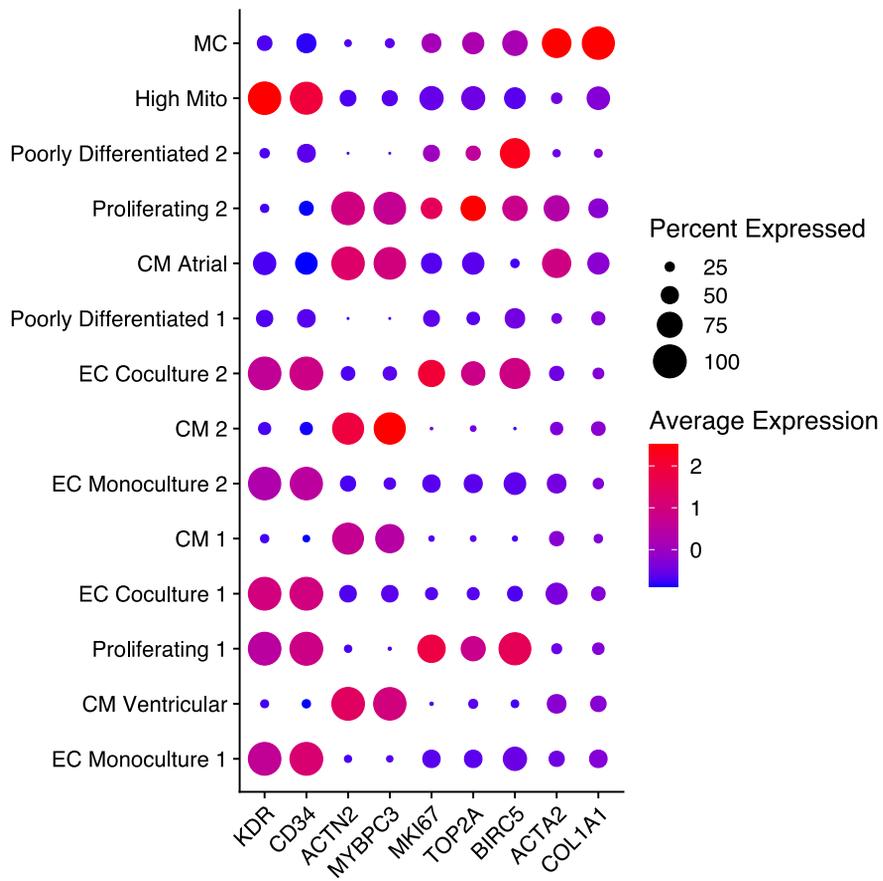
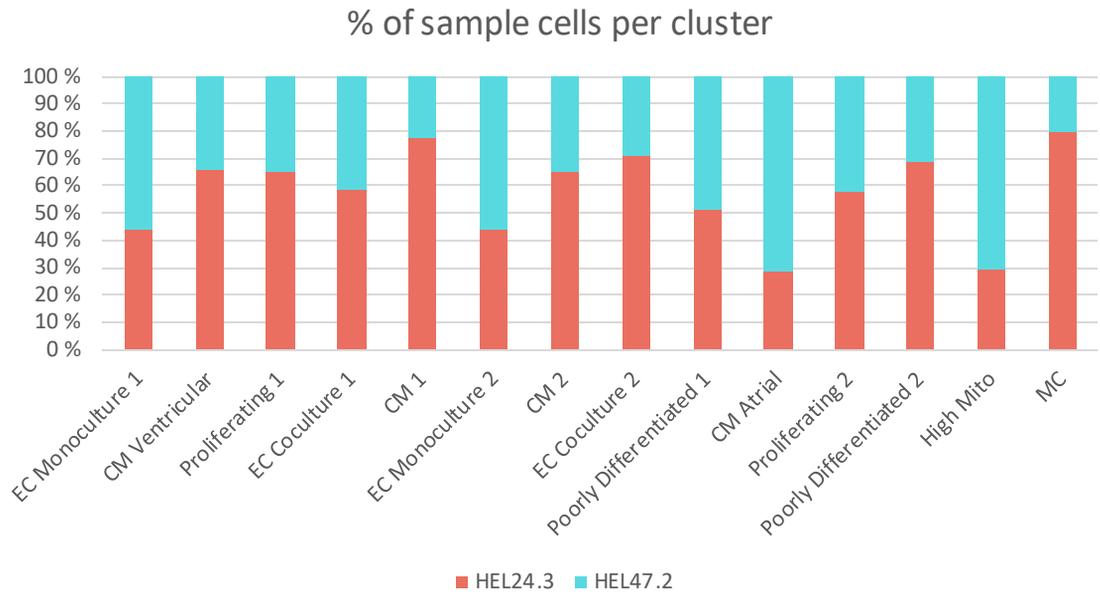


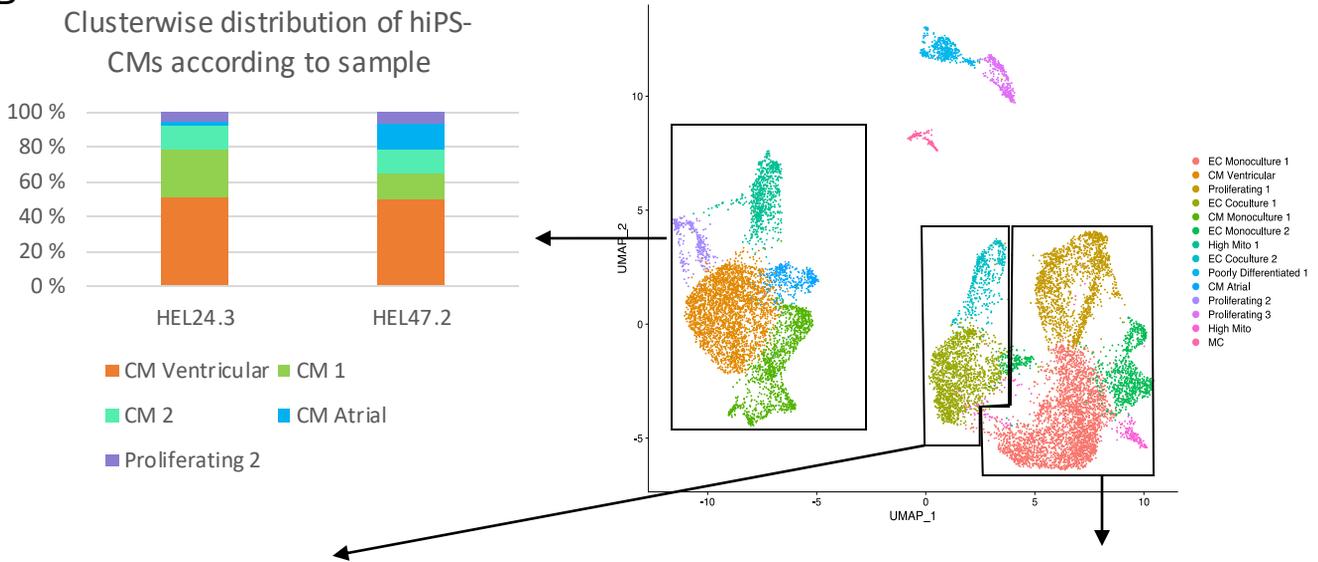
Figure S2. EC, CM, cell cycle and mesenchymal cell gene expression in the defined clusters. All scRNAseq data presented in the figure consists combined data from HEL47.2 and HEL24.3 lines.

Figure S3

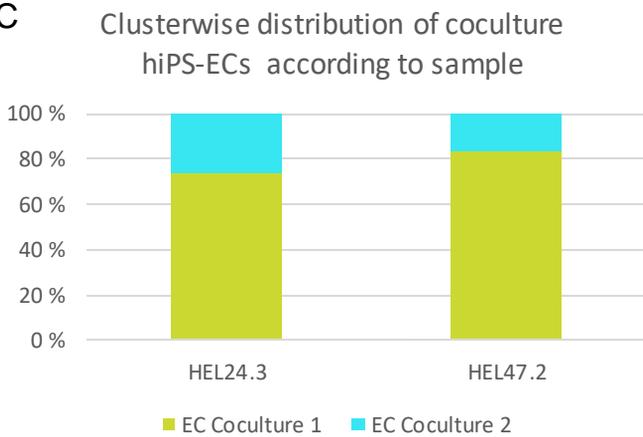
A



B



C



D

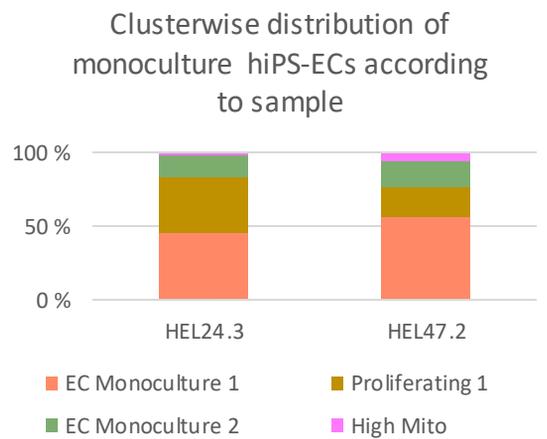


Figure S3. Comparison of hiPS transcriptomics between HEL47.2 and HEL24.3 lines. **A)** Samplewise distribution of cells per cluster. Distribution of cells per cell type cluster according to sample in **B)** hiPS-CM, **C)** hiPS-EC in coculture, **D)** hiPS-EC in monoculture.

Figure S4

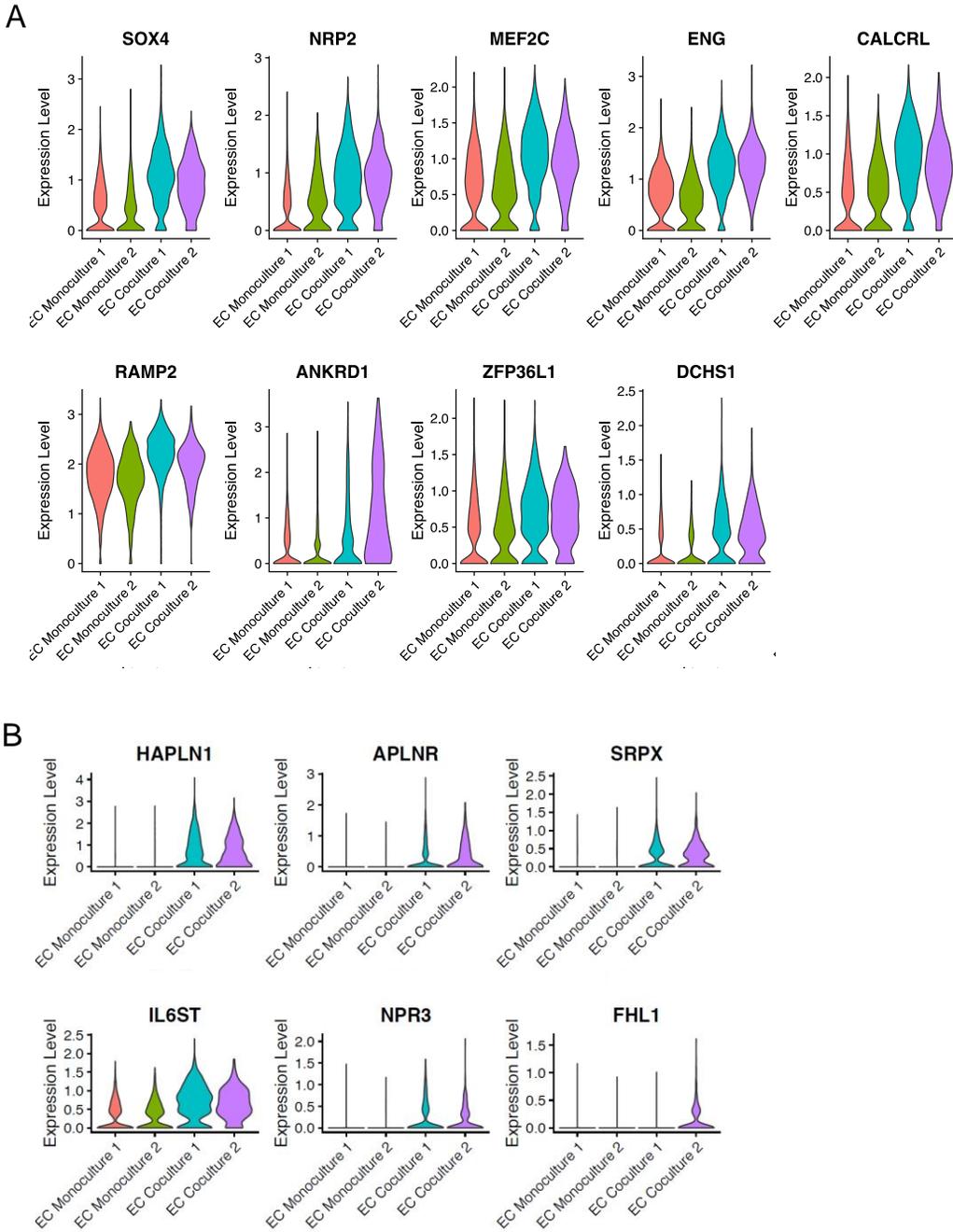


Figure S4. Coculture vs. monoculture hiPS-EC expression of **A)** genes related to heart development, **B)** cardiac EC genes. All scRNAseq data presented in the figure consists of combined data from HEL47.2 and HEL24.3 lines.

Figure S5

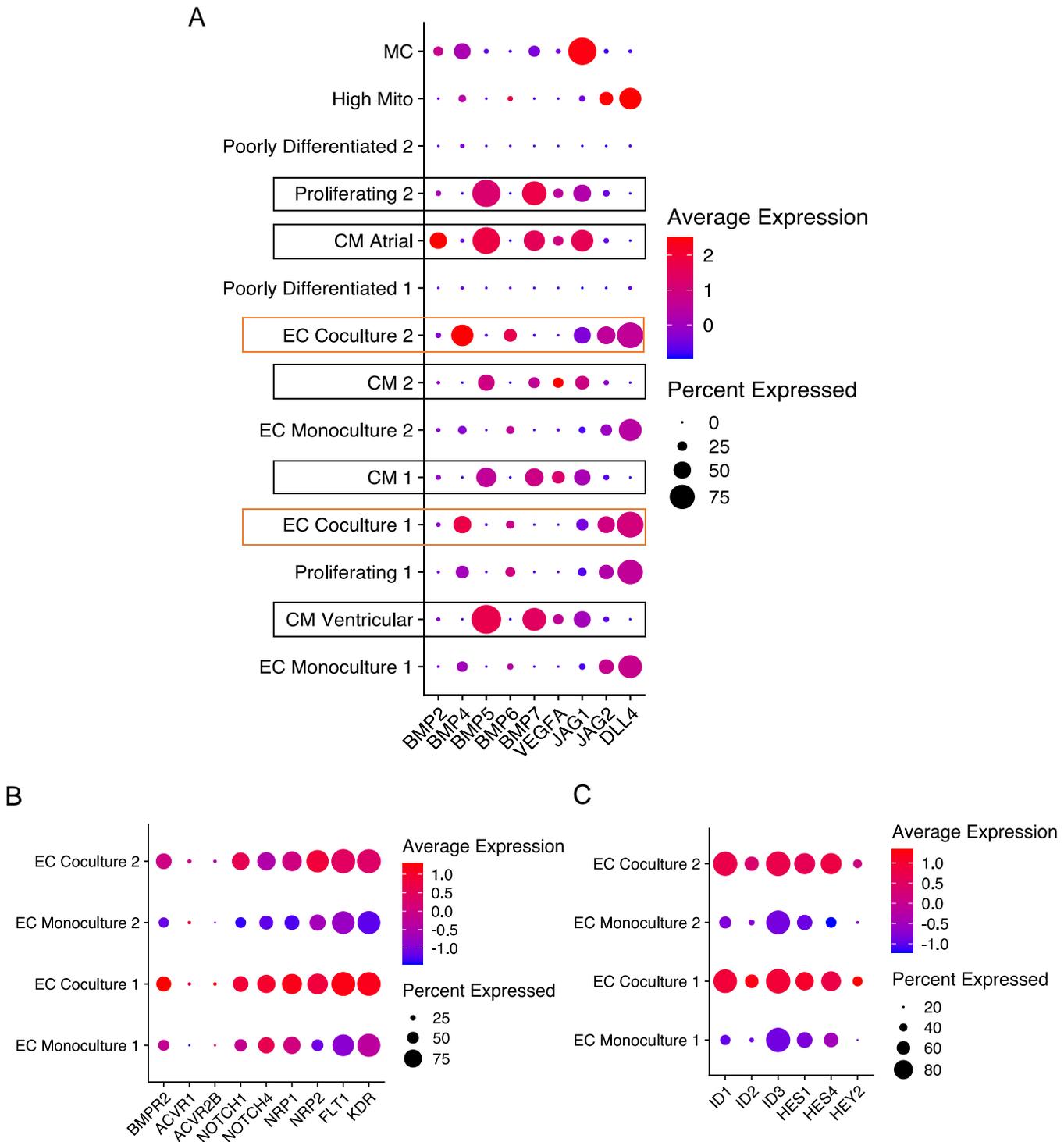


Figure S5. Changes in the expression of BMP, Notch and VEGF pathway genes. **A)** Clusterwise expression of BMP-, Notch-, and VEGF -pathway ligands included in TOP 20 hiPS-CM sender gene list and other BMP- and Notch-ligands that had higher expression in coculture hiPS-ECs. hiPS-CM and coculture hiPS-EC clusters are highlighted. Clusterwise expression of **B)** BMP-, Notch-, and VEGF -pathway receptors and **C)** BMP- and Notch -pathway targets in monoculture and coculture hiPS-ECs. All scRNAseq data presented in the figure consists of combined data from HEL47.2 and HEL24.3 lines.

Figure S6

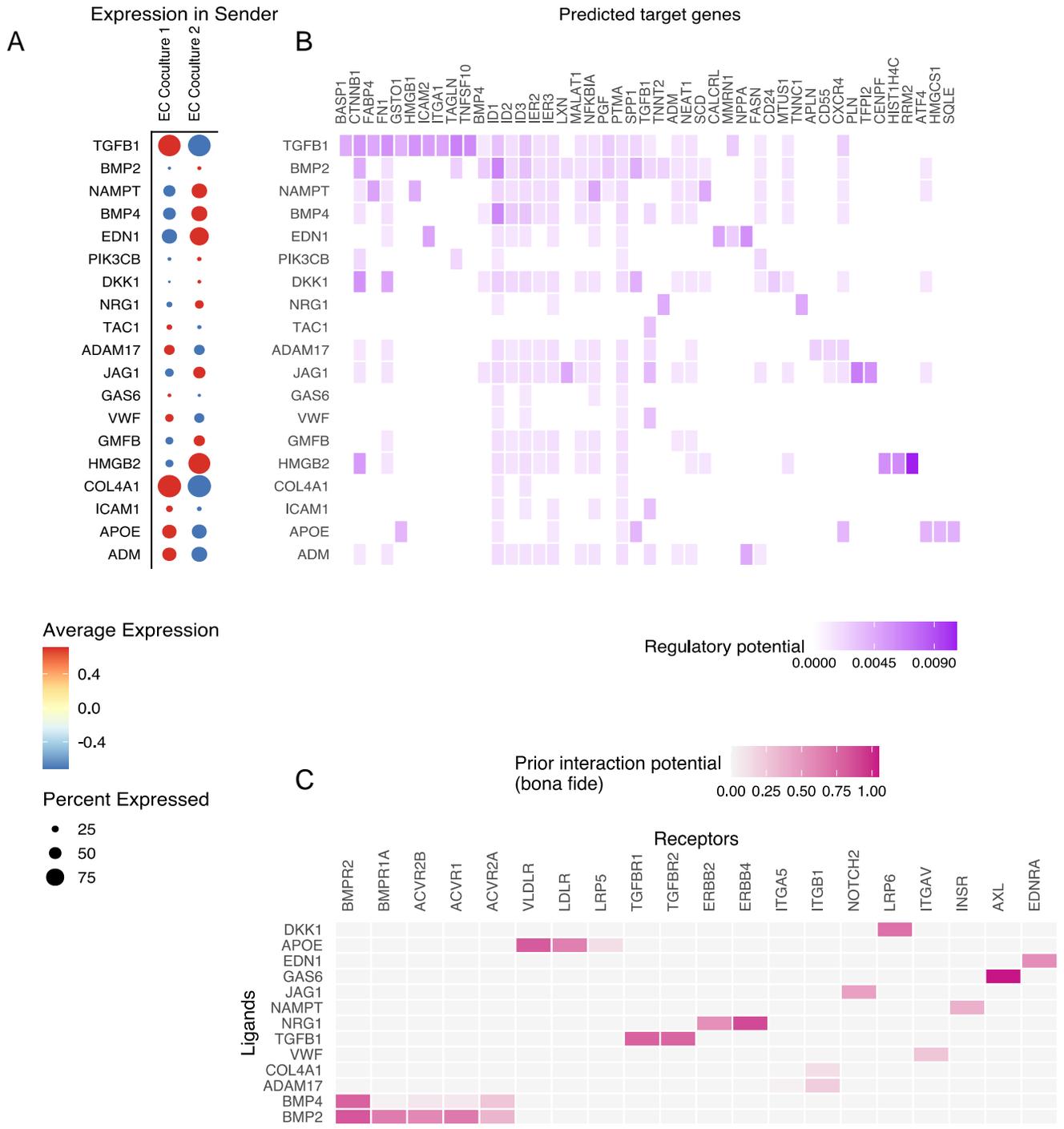


Figure S6. Ligand-target gene analysis. **A)** Expression of top ligands in sender cells (hiPS-EC). **B)** Ligand-target matrix denoting the regulatory potential between sender (hiPS-EC) ligands and predicted receiver (hiPS-CM) target genes. **C)** Ligand-receptor interactions between ligands from hiPS-ECs and receptors expressed in hiPS-CMs. All data presented in the figure consists of combined data from HEL47.2 and HEL24.3 lines.

Figure S7

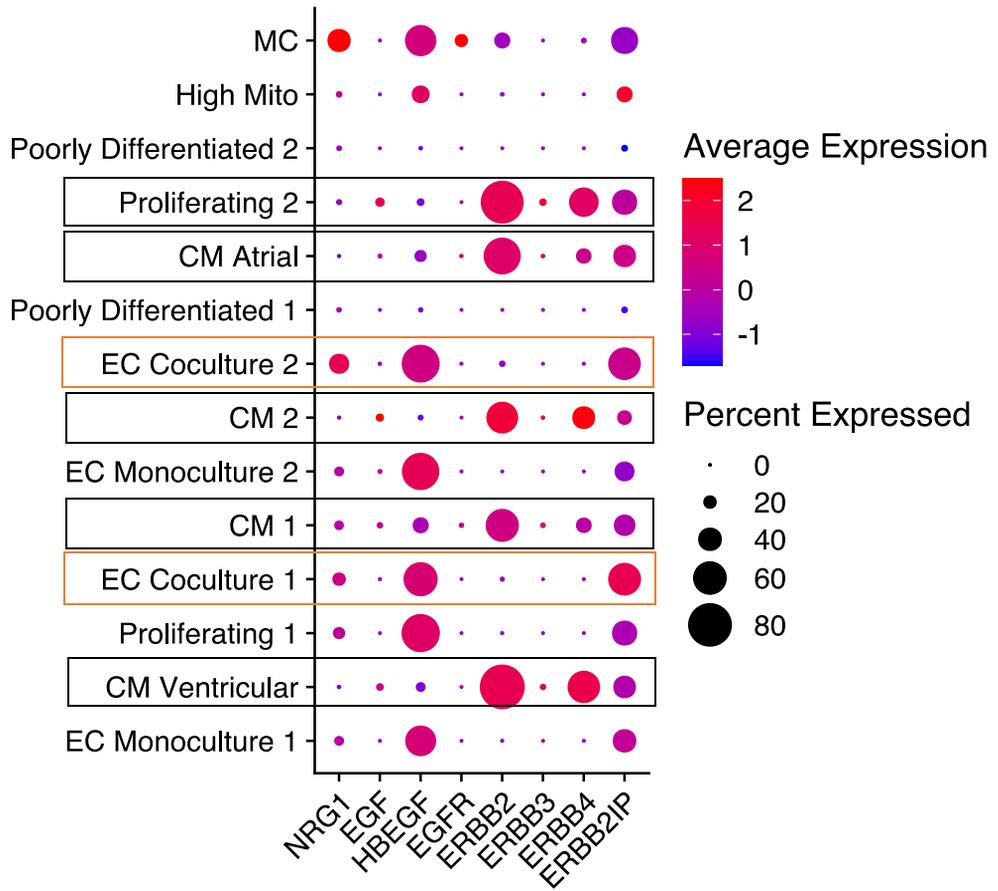


Figure S7. Clusterwise expression of ErbB-pathway ligands and receptors. hiPS-CM and co-culture hiPS-EC clusters are highlighted. All scRNAseq data presented in the figure consists of combined data from HEL47.2 and HEL24.3 lines.

Figure S8

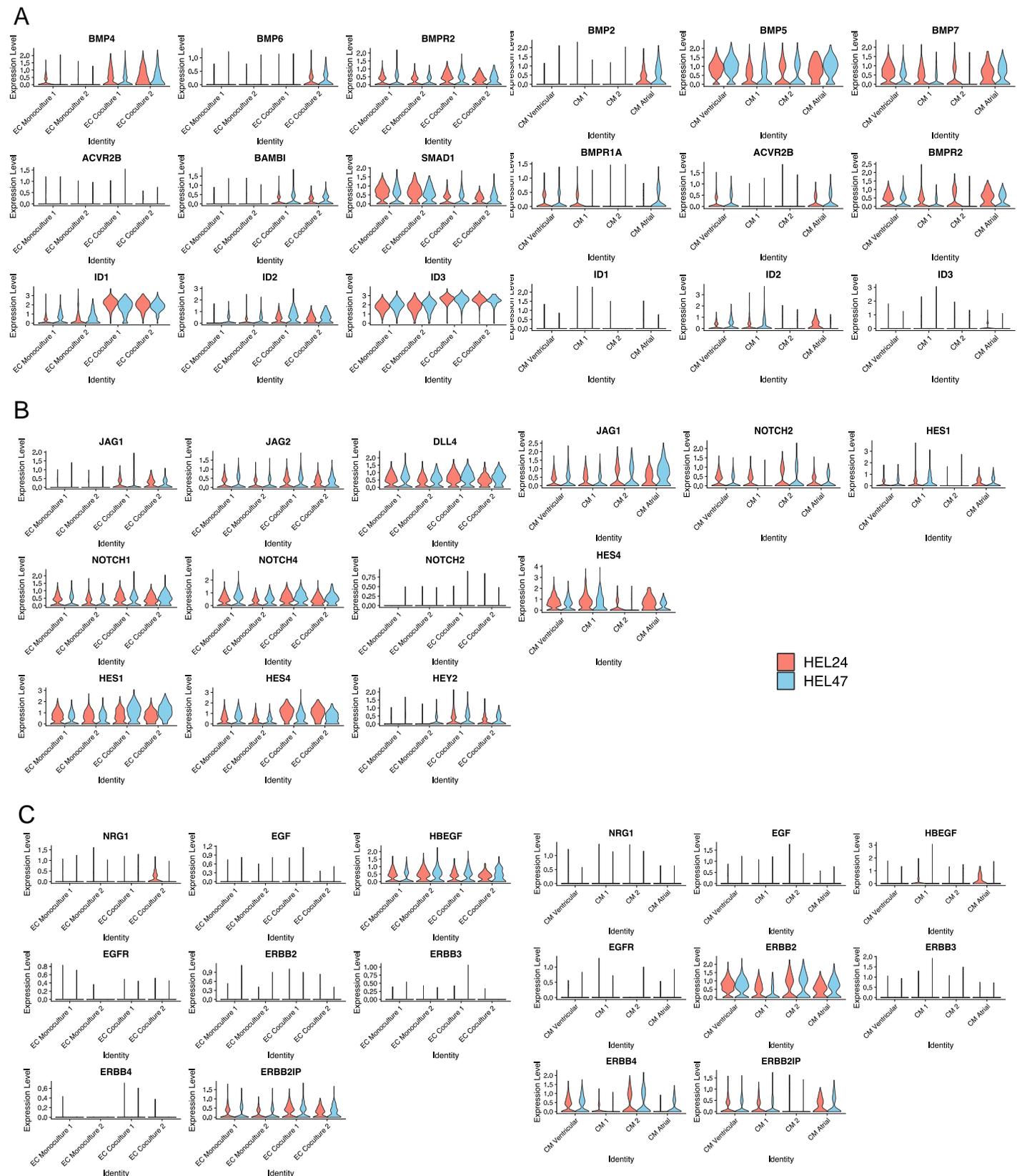


Figure S8. Clusterwise expression of **A)** BMP-, **B)** Notch-, and **C)** ErbB-pathway genes presented separately in the HEL47.2 and HEL24.3 lines.

Figure S9

