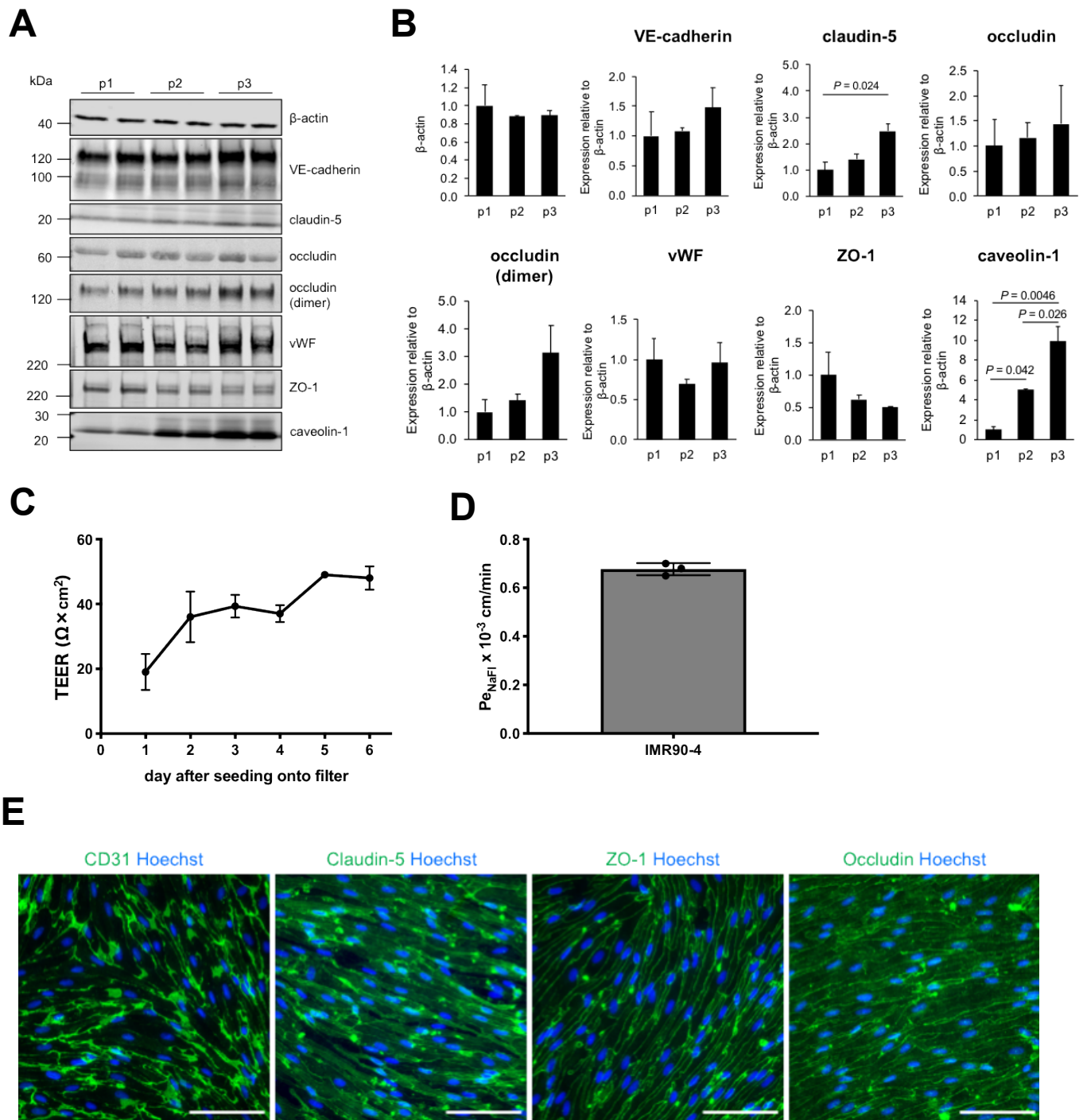


Supplementary Figure 3



Supplementary Figure 3. Western blot analysis of naive ECs and EECM-BMEC-like cells and barrier characteristics of EECM-BMEC-like cells from an hiPSC line originating from fibroblasts.

(A) Western blots of naive EC (passage 1, p1) and EECM-BMEC-like cells (passage 2 and 3, p2 and p3) probed for β -actin, VE-cadherin, claudin-5, occludin, vWF, ZO-1, and caveolin-1. (B) Quantification of Western blot band intensity. Data are shown as the mean \pm SD of two wells from one differentiation of the IMR90-4 hiPSC line. Statistical analysis: one-way ANOVA followed by Tukey's multiple comparison test. (C, D) TEER (C) and permeability to sodium fluorescein (D) of EECM-BMEC-like cell monolayers from IMR90-4 hiPSC line is shown. EECM-BMEC-like cells were seeded onto 0.4 μm pore size Transwell filters as monoculture and TEER were measured over 6 days and PeNaFl was measured at day 6. (C) Plotted data are mean TEER values \pm SD. (D) Bars show the mean permeability coefficients (Pe) \pm SD. (E) Immunofluorescence images of EECM-BMEC-like cells from IMR90-4 hiPSC line grown on 0.4 μm pore Transwell filters. Cell junctions were stained for CD31 (green), claudin-5 (green), zonula occludens 1 (ZO-1, green), occludin (green) and nuclei were stained with Hoechst (blue). Scale bars = 100 μm .