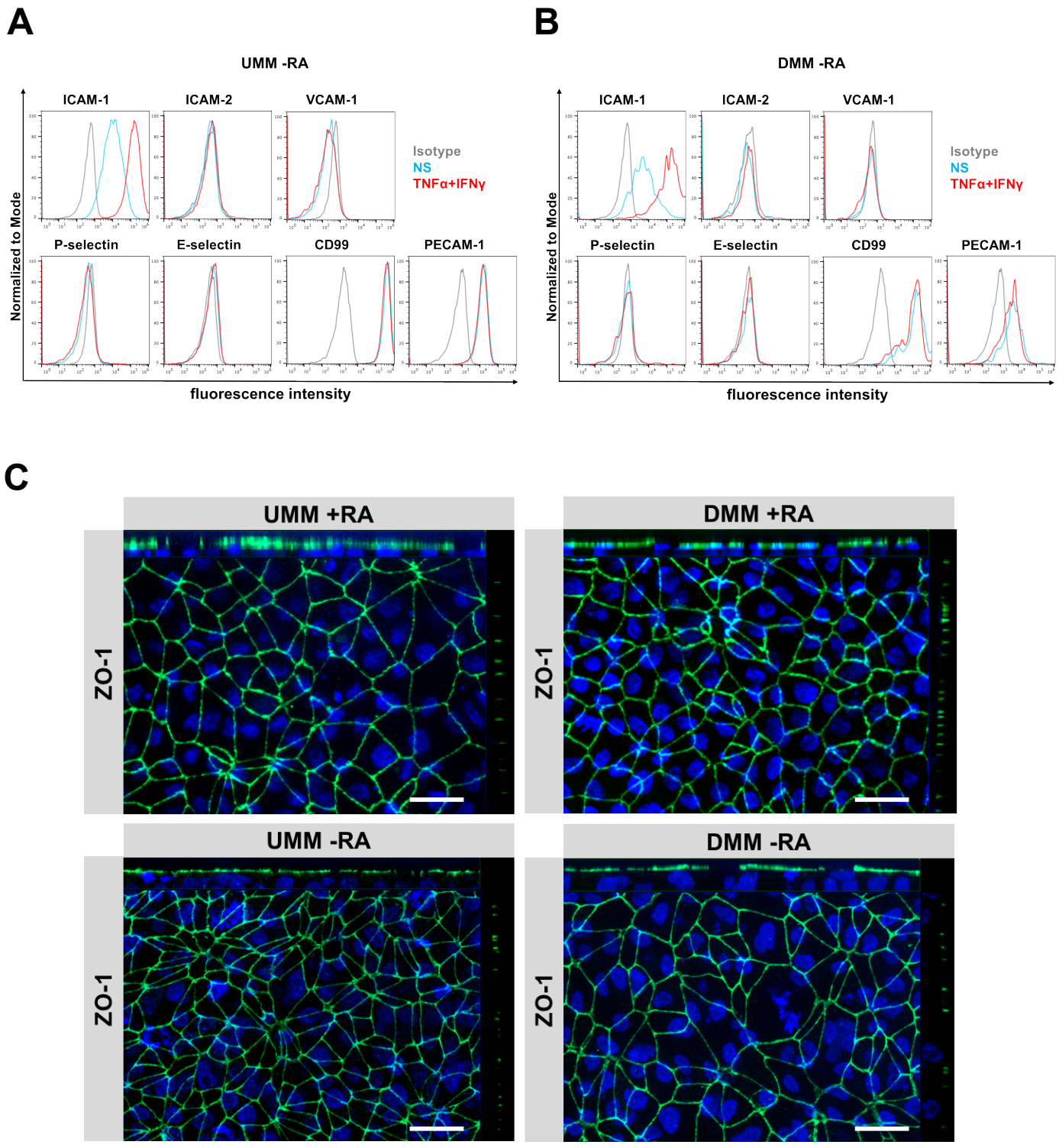
Supplementary Figure 1



Supplementary Figure 1. Adhesion molecule phenotype and morphology of BMEC-like cells differentiated by unconditioned medium method (UMM) or defined medium method (DMM) from additional hiPSC clone.
Cell surface staining of BMEC-like cells differentiated by UMM (A) or DMM (B) in the absence of RA for the adhesion molecules ICAM-1, ICAM-2, VCAM-1, P-selectin, E-selectin, CD99, and PECAM-1 was analyzed by flow cytometry. Isotype control, non-stimulated (NS), and 16 h pro-inflammatory cytokine-stimulated condition (10 ng/mL TNF-α + 200 IU/mL IFN-γ) are represented respectively in grey, blue, and red lines in a histogram overlay. Representative data from donor 3 are shown. (C) Immunofluorescence images of UMM- or DMM-differentiated BMEC-like cells grown on 0.4 µm pore Transwell filters. Cell junctions were stained for zonula occludens 1 (ZO-1, green) and nuclei were stained with DAPI (blue). The bottom of each panel shows a maximum-intensity projection through the z-axis. In the top (xz) and right (yz) side images of each panel the apical side of the BMEC-like cells is oriented, respectively, towards the top and the right side. Representative data from donor 2 are shown. Each staining is representative of at least 3 independent differentiations performed on 3 distinct filters. Scale bars = 20 µm.