

Supplementary Figures

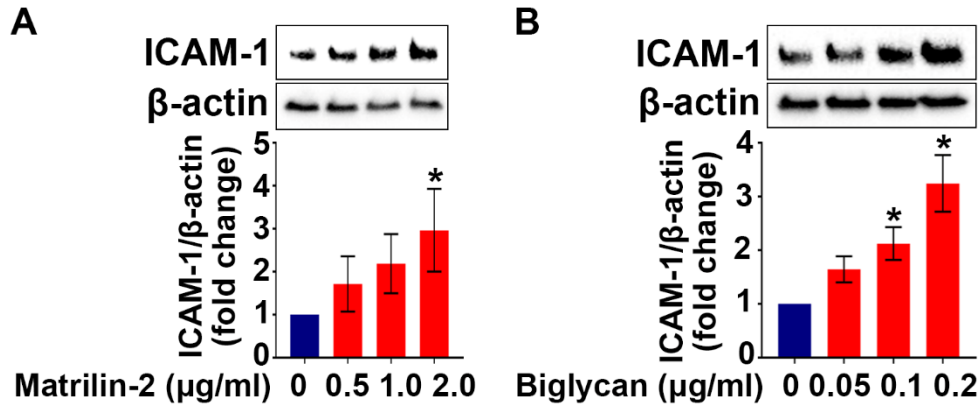


Figure S1. Soluble ECM proteins induce AVICs inflammatory response. Human AVICs were stimulated with recombinant matrilin-2 or biglycan with different concentrations. Values are mean ± SEM, n = 3 donors. *P<0.05 vs. control.

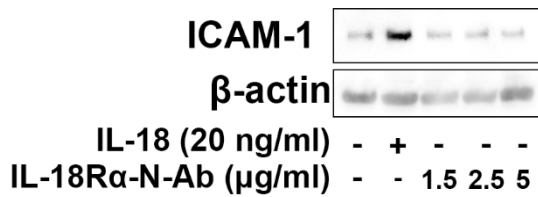


Figure S2. Neutralization of IL-18Rα suppresses the upregulation of ICAM-1 expression induced by IL-18. Human AVICs were stimulated with IL-18Rα neutralizing antibodies prior to recombinant IL-18 treatment. Neutralization of IL-18Rα markedly reduced ICAM-1 level in human AVICs following IL-18 treatment. n = 2 donors.

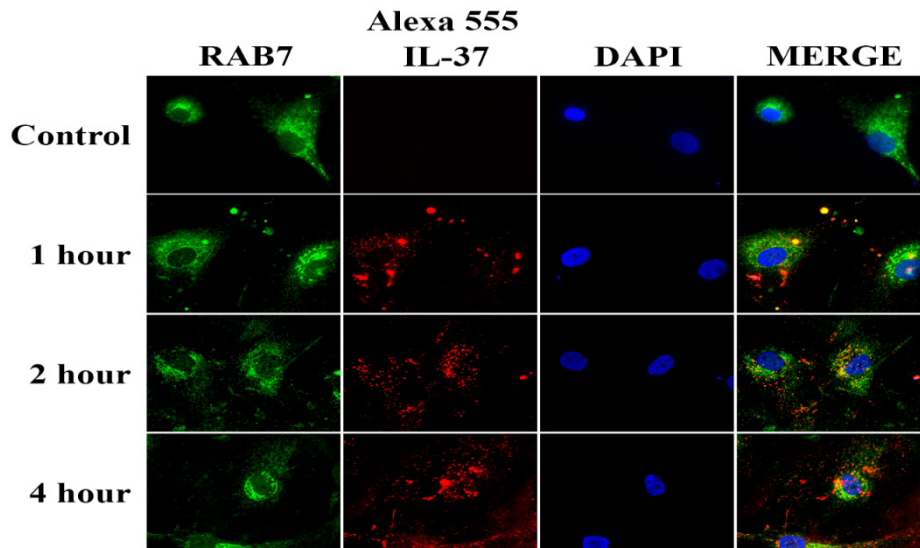


Figure S3. The internalization of recombinant IL-37 into human AVICs is associated with late endosomes. Cells were treated with tagged IL-37 protein. Representative images of immunofluorescence staining show that IL-37 did not colocalize with Rab7 (late endosomes). Red: Tagged IL-37; Green: Rab7; and Blue: DAPI. Original magnification, 63x objective.

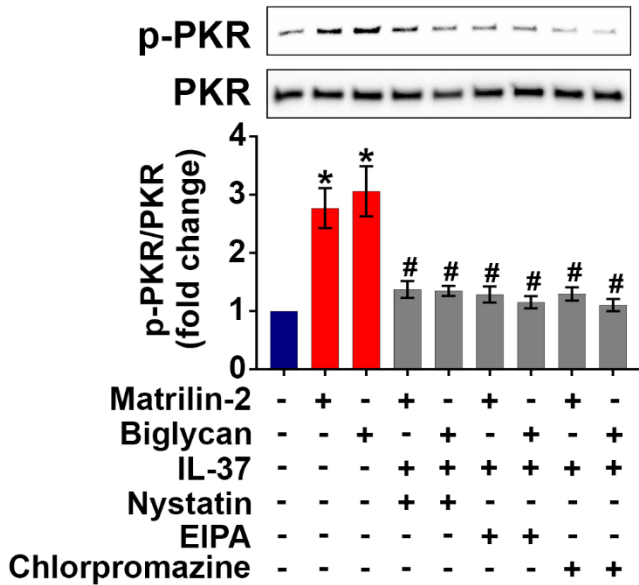


Figure S4. Endocytosis inhibitors do not attenuate the inhibitory effects of IL-37 on PKR phosphorylation. Human AVICs were treated with pharmacological inhibitors of endocytosis prior to recombinant IL-37 and matrilin-2 or biglycan treatment. n = 4 donors. * $P < 0.05$ vs. control. # $P < 0.05$ vs. cells treated with matrilin-2 alone or biglycan alone.

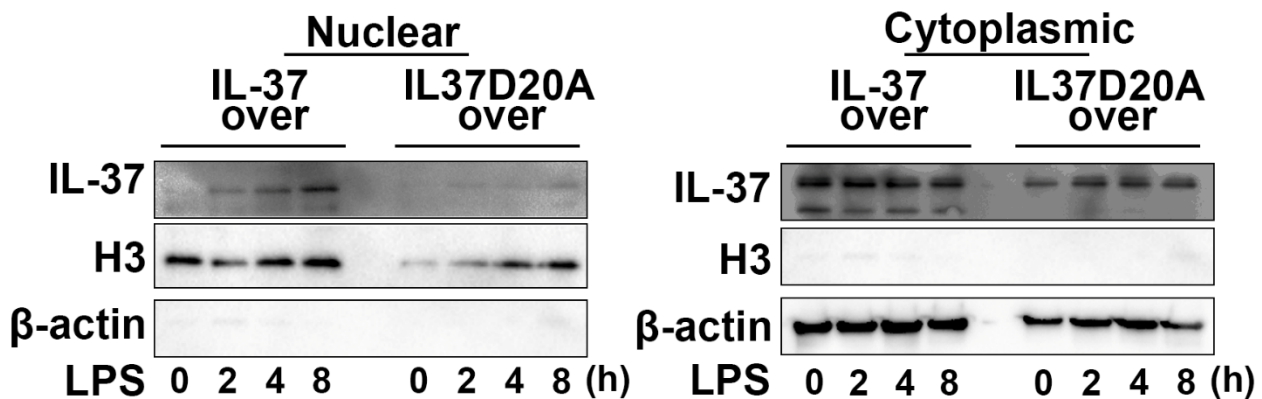


Figure S5. IL-37 expression is detected in nuclear fraction of AVICs overexpressing IL-37 but not in IL-37D20A. Nuclear and cytoplasmic fractions from human AVICs were extracted after cells overexpressed with plasmid EGFP-IL-37 and EGFP-IL-37D20A and treated with LPS (200 ng/mL). n = 2 donors.

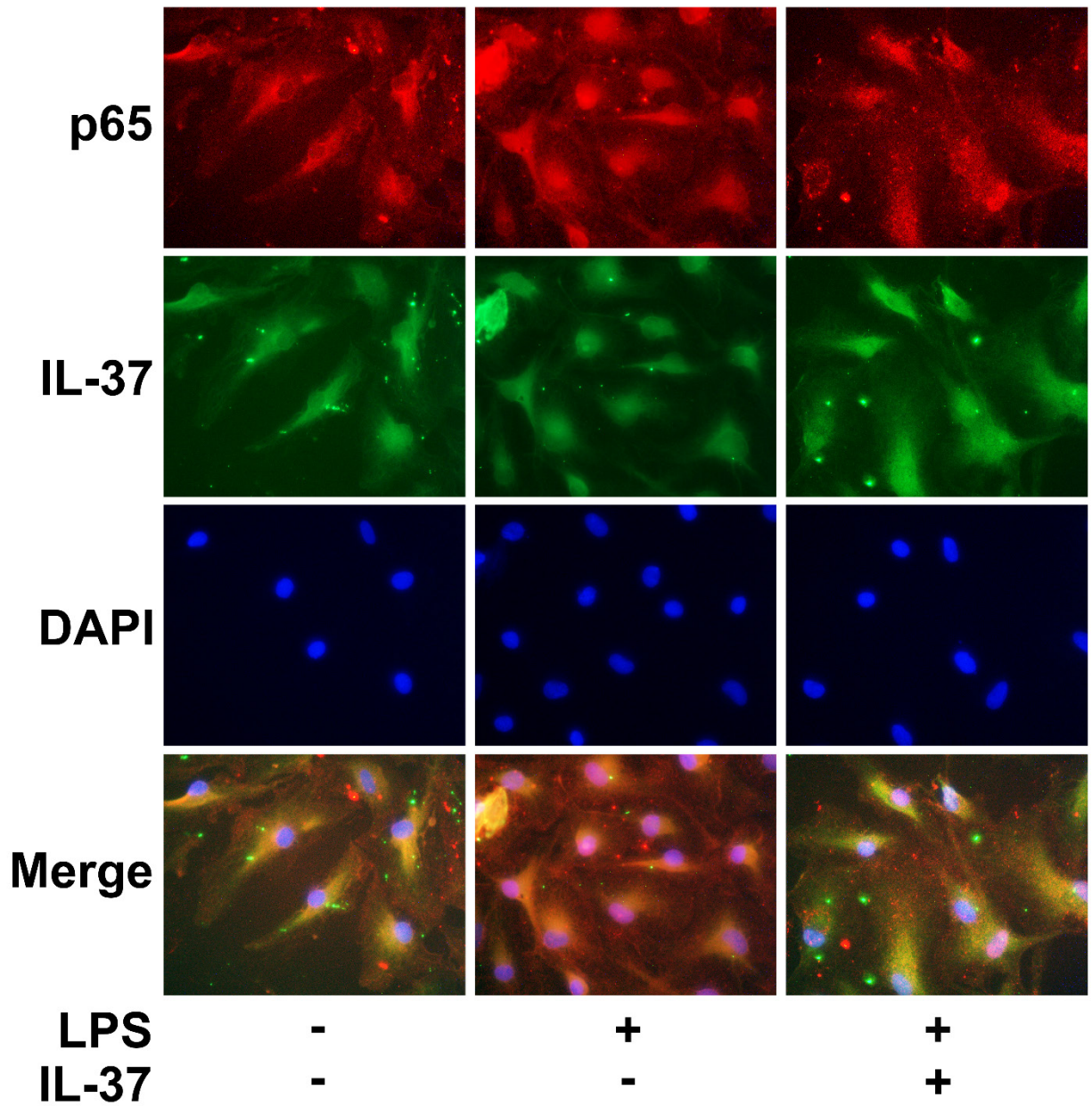


Figure S6. Treatment with recombinant IL-37 reduces NF- κ B intranuclear translocation. Human AVICs were pre-treated with recombinant IL-37 at a concentration of 1 ng/mL for 24 hours followed by an exposure to Toll-like receptor 4 agonist LPS (200 ng/mL) for 4 hours. Representative images show that IL-37 translocates to the nucleus in response to LPS stimulation in cells with or without recombinant IL-37. Treatment with recombinant IL-37 increases nuclear level of IL-37 and attenuates NF- κ B p65 intranuclear translocation in cells exposed to LPS. Original magnification, 40x objective.