

P005 The adhesion molecule ICAM-2 regulates contact inhibition in endothelial cells (EC)
Valerie Amsellem¹, Nicola Dryden¹, Graeme M. Birdsey¹, Dorian O. Haskard¹, Justin Mason¹ and Anna M. Randi¹

¹NHLI Imperial College London, Hammersmith Hospital, BHF Cardiovascular Science, London, UK

Endothelial junctions play a key role in important processes, including contact inhibition and angiogenesis. ICAM-2, constitutively expressed at endothelial junctions, mediates leukocyte recruitment and angiogenesis. The ICAM-2 intracellular tail binds ERMs and α -actinin. We generated an ICAM-2 deficient EC line (Δ IC2) from mouse cardiac EC, isolated from ICAM-2 $-/-$ mice by positive selection and immortalized with polyoma middle T. We also generated an over-expressing line (Δ IC2-IC2), by infecting the Δ IC2 cell line with a retroviral vector encoding for full-length mouse ICAM-2, and two mutant ICAM-2 lines, one lacking the intracellular tail (Δ IC2-IC2 Δ TL) and one where the ERM binding sites have been mutated (Δ IC2-IC2 Δ ERM). The Δ IC2 EC line shows defective *in vitro* angiogenesis on Matrigel, decreased cell migration and Rac activity; over-expression of ICAM-2 (Δ IC2-IC2) reverses these phenotypes. Interestingly, Δ IC2-IC2 EC form a cobblestone monolayer at confluence and are contact-inhibited whilst the Δ IC2 cells overlap, suggesting that ICAM-2 is required for contact-dependent inhibition of cell growth. This involves signalling via ERMs, since both mutants (Δ IC2-IC2 Δ TL) and (Δ IC2-IC2 Δ ERM) are not contact-inhibited. Contact inhibition can be restored by over-expressing constitutively active Rac. Thus ICAM-2 regulates EC growth and contact inhibition via signalling pathways involving ERMs and Rac.