



Supplement Figure 1: BMP9 increases cell surface E-selectin expression in HPAECs. HPAECs were serum-restricted in M199/0.1% FBS overnight and then incubated with BMP9 (10ng/ml) in M199/0.1% (0.1% FBS) for 6h. As a positive control, HPAECs were incubated with TNF- α (5ng/ml), a known stimulus of E-selectin expression in endothelial cells (1). At the end of the incubation period, cells were detached with Accutase (PAA Laboratories Ltd., Yeovil, Somerset, UK) and washed twice with PBS/0.5% BSA. HPAECs were then aliquoted at 1×10^5 cells/20 μ l and incubated with 10 μ l FITC-conjugated mouse anti-human E-selectin antibody (AbD Serotec, Kidlington, Oxford, UK) for 1h at 4 $^{\circ}$ C. Cells were washed twice with PBS/0.5% BSA and resuspended in 0.5ml PBS/0.5% BSA containing 1 μ g/ml propidium iodide. Cells were analysed for FITC and propidium iodide fluorescence by flow cytometry. As expected, TNF- α increased cell surface E-selectin expression, determined as increased fluorescence on the FL1-H channel represented as a shift into the marked region (R1). BMP9 also induced an increase in cell surface E-selectin expression, demonstrated by a shift of fluorescence into R1. Data are representative of 2 separate experiments.

1. Read, M. A., Whitley, M. Z., Gupta, S., Pierce, J. W., Best, J., Davis, R. J., and Collins, T. (1997) *J. Biol. Chem.* **272**, 2753-2761