



Supplement Figure 2: The pSmad3 antibody cross-reacts with pSmad1, but the BMP9-induced band of lower molecular mass is not pSmad3. (A) Protein samples from HPAECs treated with BMP9, BMP2, BMP4, BMP6 or TGF β ₁ were immunoblotted for phospho-Smad3 followed by reprobing with Smad3 antibody. The phosphorylated band of higher molecular mass corresponds to Smad1 and is distinct from the Smad3 protein. (B) Protein samples from HPAECs transfected with Smad2 or Smad3 siRNA were immunoblotted for phospho-Smad3 followed by reprobing with Smad3 antibody. The phosphorylated band of higher molecular mass corresponds to Smad1 and is distinct from the Smad3 protein, which is absent in cells transfected with siSmad3. (C) HMEC-1 cells were transfected with siSmad3 or siCP (10nM each) using Dharmafect1 (DH1). After 28h, cells were serum-restricted for 16h, followed by treatment with BMP9 (1ng/ml) or TGF β ₁ (5ng/ml) in 0.1% for 1h. Immunoblotting was performed with antibodies against phospho-Smad3 or Smad3. Blots are representative of 2 repeats.