



Supplementary Figure S1 Wnt3a/EGF forms tubular structures of IEC6 cells in 3D culture. **(A)** IEC6 cells were cultured with Wnt5a, Wnt5a/EGF, or Wnt5a/HGF for 72 h in 3D culture. The representative structures were photographed using phase contrast microscopy (top panels). The cells were stained with anti- β -catenin antibody and phalloidin (bottom panels). **(B)** IEC6 cells were cultured with EGF for 60 h in 3D culture and stained with anti- β -catenin antibody and phalloidin. White boxes show enlarged images. **(C)** IEC6 cells were cultured with Wnt3a/EGF for 60 h and stained with anti-aPKC, anti-phospho-ezrin, and anti-E-cadherin antibodies and phalloidin. **(D)** IEC6 cells were cultured with Wnt3a/EGF for 60 h in 3D culture and stained with anti- β -catenin antibody and phalloidin. The number of extended structures were counted as four. **(E)** IEC6 cells were cultured with Wnt3a/EGF or Wnt3a/HGF for 72 h in 3D culture. The number of extended structures from multicellular trunks were counted (n=30). Results are shown as the mean \pm SE from three independent experiments. *, $P < 0.01$. Scale bars in **(A)**, 200 μ m (top panels) and 20 μ m (bottom panels); in **(B)**, 20 μ m; in **(C and D)**, 50 μ m.