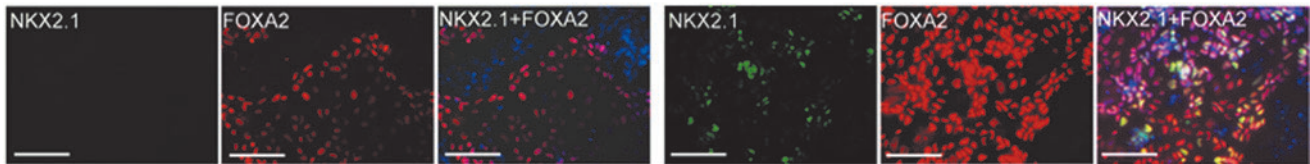
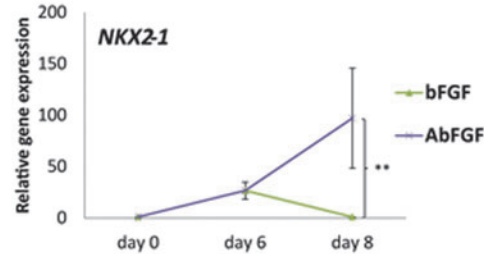
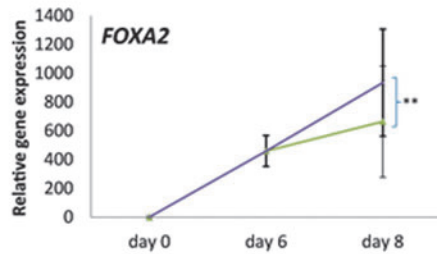
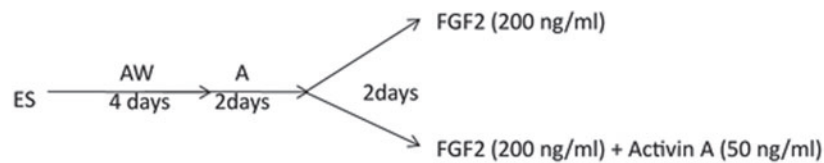
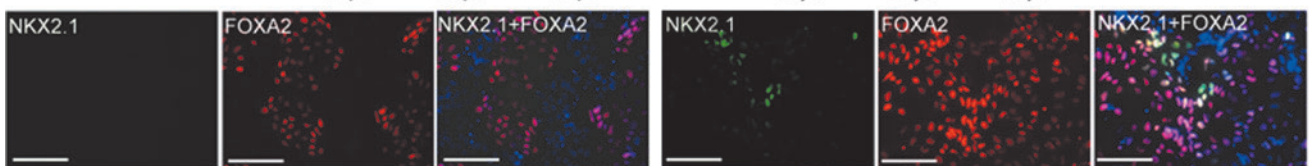
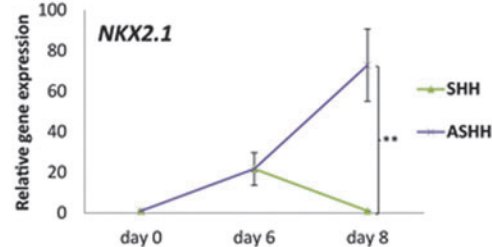
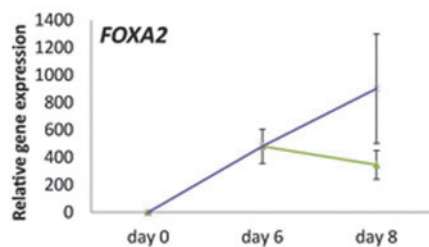
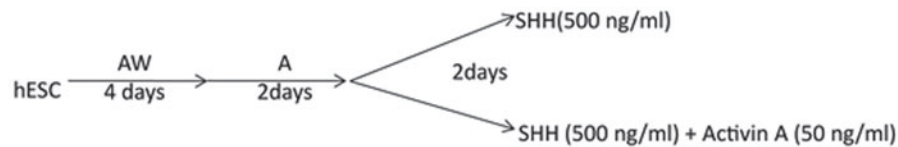
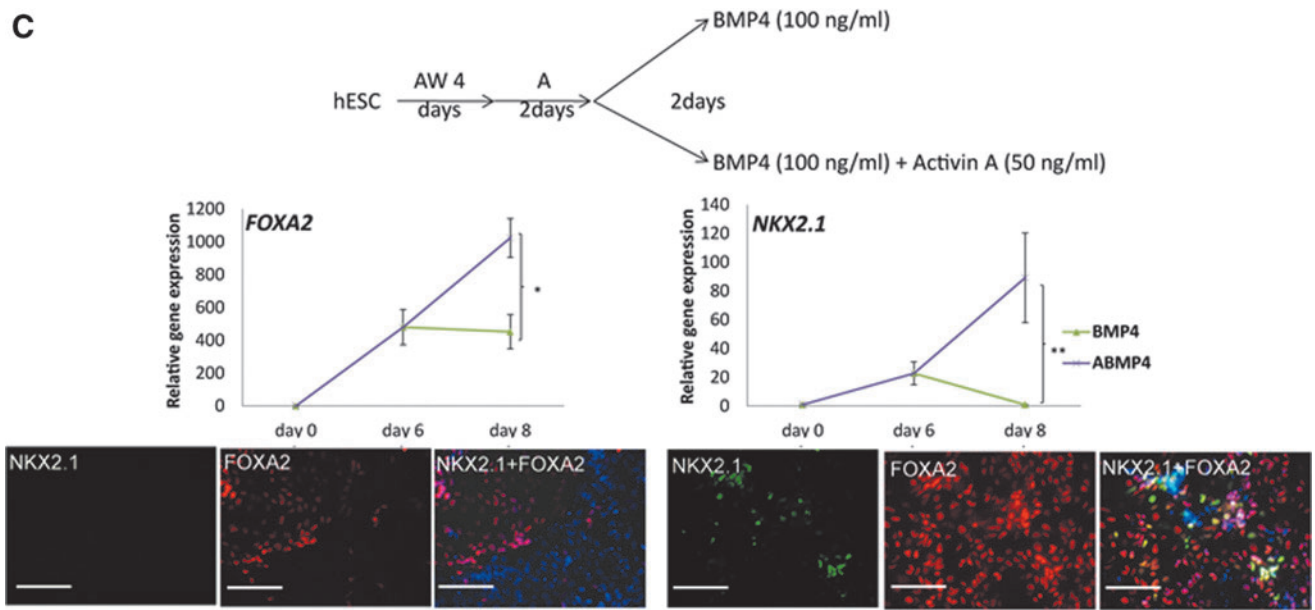
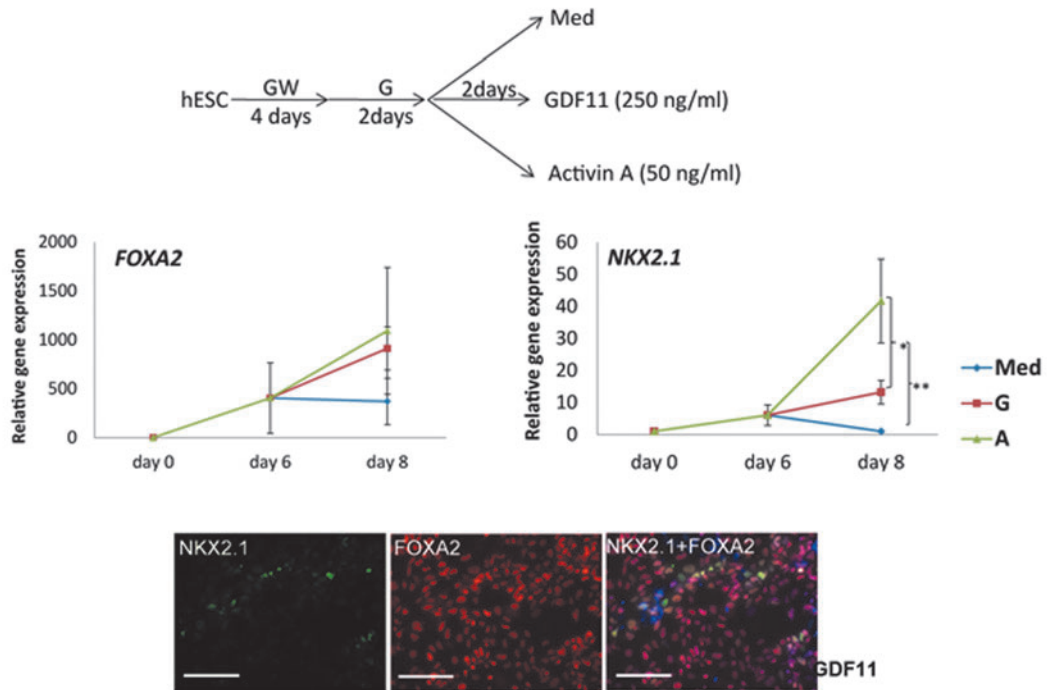


A**B**

SUPPLEMENTARY FIG. S2. Neither Fibroblast growth factor-2 (FGF2), sonic hedgehog (SHH) or BMP4 induce NKX2-1; while GDF11 has similar NKX2-1 induction capacity as Activin-A. **(A–C)** NKX2-1 and FOXA2 expression in hESC-H9 progeny cultured for 4 days with Activin-A + Wnt3a, 2 days with Activin-A followed by FGF2, SHH or BMP4. All data are expressed as mean \pm SEM of different experiments. (* P < 0.05; ** P < 0.01). Scale bar, 100 μ m. **(A)** 2 additional days with FGF2 alone or combined with Activin-A (n = 6): Percentage FOXA2 positive cells: FGF2 = 76.6% \pm 16.6%, AFGF2: 84.1% \pm 7.4%; Percentage NKX2-1 positive cells: 0% in FGF2 condition, and 10% \pm 9.4% in AFGF2. **(B)** 2 additional days of SHH alone or combined with Activin-A (n = 4): Percentage FOXA2 positive cells: SHH = 64.0% \pm 11.3%, ASHH: 73.2% \pm 14.8%; Percentage NKX2-1 positive cells: 0% in SHH condition, and 10.1% \pm 9% in ASHH. **(C)** 2 additional days of BMP4 alone combined with Activin-A (n = 5): Percentage FOXA2 positive cells: BMP = 70.1% \pm 10.1%, ABMP: 71.1% \pm 18.2%; Percentage NKX2-1 positive cells: 0% in BMP condition, 9.9% \pm 7.8% in ABMP. **(D)** NKX2-1 and FOXA2 expression in hESC-H9 progeny cultured for 4 days with GDF11 + Wnt3a, and 4 days of GDF11 alone (n = 3). Scale bar, 100 μ m. Percentage FOXA2 positive cells 72.4% \pm 15.8%; Percentage NKX2-1 positive cells: 4.3% \pm 2.6%. ASHH, Activin A + SHH; ABMP, Activin A + BMP.

C**D**

SUPPLEMENTARY FIG. S2. (Continued).