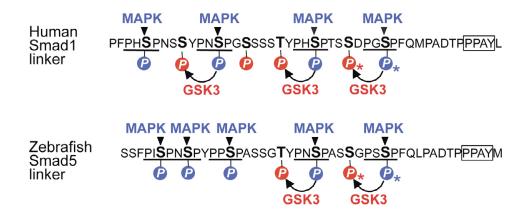
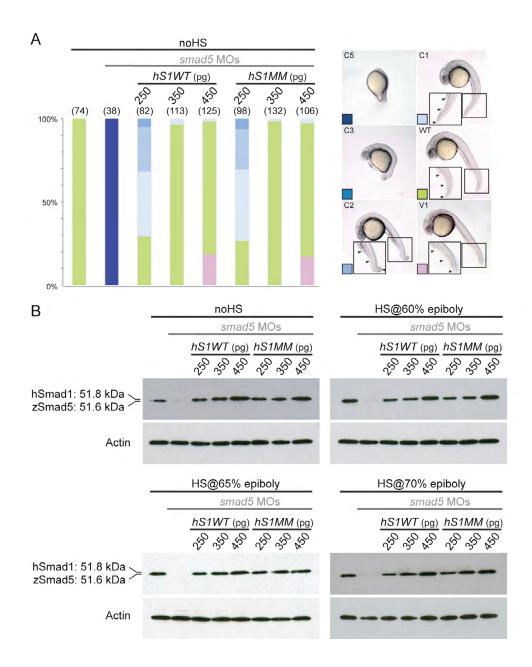


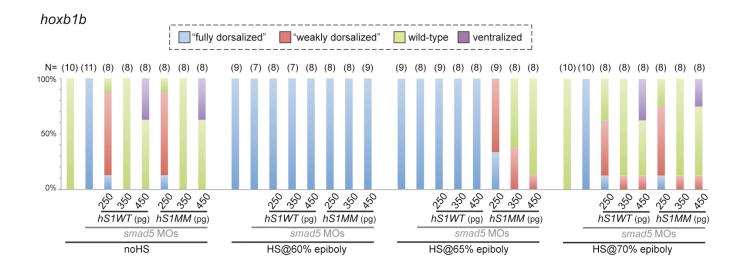
Fig. S1. Anteriorly expanded pax8, caused by activation of RA signaling, is patterned by BMP signaling with the same temporal dynamics as the normal posterior domain. (A-E) Expression of pax8 (arrowheads) in wild type (A, n=42/42) and in Tg(hsp70:chd) heat shocked at sphere (B, n=38/38), 40% epiboly (C, n=28/31), 50% epiboly (D, n=31/35) and shield (E, n=30/31) stage. (F-J) pax8 expression in RA-treated wild type (F, n=38/39) and RA-treated and heat shocked Tg(hsp70:chd) at sphere (G, n=30/30), 40% epiboly (H, n=31/35), 50% epiboly (I, n=32/34) and shield (J, n=37/38) stage. Lateral views, dorsal to right with anterior to top, except for insets in A and F, which are dorsal views with anterior to top. All are at the 6-somite stage.



**Fig. S2. FGF/MAPK and Wnt/GSK3 phosphorylation sites in the zebrafish Smad5 linker region.** Zebrafish Smad5 contains MAPK (blue) and GSK3 (red) phosphorylation sites in its linker region. The box indicates the PPAY binding site of Smurf1 and the asterisks indicate antibody recognition sites for P-Smad1/5<sup>MAPK</sup> and P-Smad1/5<sup>GSK3</sup>.



**Fig. S3.** *hSmad1WT* and *hSmad1MM* mRNAs similarly rescue Smad5-deficient dorsalized embryos and hSmad1WT and hSmad1MM are equally stable in the injected embryos. (A) Percentage of embryos that show severely (C5), moderately (C3) and mildly (C1/C2) dorsalized, wild-type (WT) and V1 (diminished eye size and increased ventral tail fin tissue) phenotypes in control and *smad5* MO-injected Tg(*hsp70:chd*) embryos without heat shock, also injected with either *hSmad1WT* mRNA (250, 350 or 450 pg) or *hSmad1MM* mRNA (250, 350 or 450 pg). (B) Western blot for total Smad1/5 protein in the same embryos as shown in A. Embryos were collected at 80% epiboly stage. Control and *smad5* MO-injected Tg(*hsp70:chd*) embryos, also injected with *hSmad1WT* mRNA (250, 350 or 450 pg) and *hSmad1MM* mRNA (250, 350 or 450 pg), and either not heat shocked (noHS; upper left panel) or heat-shocked at 60% epiboly (upper right), 65% epiboly (lower left) or 70% epiboly (lower right).



**Fig. S4.** *hSmad1MM* causes precocious patterning of DV tissues. Percentage of embryos exhibiting a fully dorsalized, weakly dorsalized, wild-type or ventralized phenotype in control and *smad5* MO-injected embryos, also injected with *hSmad1WT* mRNA (250, 350 or 450 pg) and *hSmad1MM* mRNA (250, 350 or 450 pg), and with either no heat shock or heat shocked at different stages to induce expression from Tg(*hsp70:chd*).