Supplemental Figure 1

BMP4 (+) BMP4 (-)

Α



IsI1^{flox/flox}

BMP4 (-)

Β





Hoxb6Cre;IsI1^{flox/flox}





Hoxb6Cre;IsI1^{flox/flox}





Supplemental Figure 2

Isl1Cre; indicator









Supplemental Figure 1. BMP4 addition to *Isl1* mutant kidneys in organ culture. Each side of the metanephric region at E10.5 was isolated and cultured on a filter at the air/liquid interface for 24 hours, and stained with an anti-cytokeratin 8 antibody to visualize the ureteric buds. (A) BMP4 (1 ng/ml) was added to the culture medium. Four control kidneys and five *Isl1* mutant kidneys were used for each condition. (B) Affi-Gel Blue beads (Bio-Rad) soaked for 1 hour in BMP4 (5 µg/ml) or albumin (negative control) were placed close to the presumptive ectopic budding sites of the nephric ducts, as shown by dotted circles (six kidneys in each condition). Ureteric budding was impaired or accessory buds (**) were formed in the Isl1 mutants, irrespective of BMP4 treatment. Beads soaked in BMP4 (0.5 µg/ml) showed similar results (four kidneys in each group). While the indicated concentrations of BMP4 had minimal effects on the control kidneys, higher concentrations of BMP4 inhibited ureteric budding in both the controls and the Isl1 mutants. When the BMP4-soaked beads were placed on the metanephric side of the nephric duct, we observed more inhibitory effects. White arrowhead: ureteric bud tip; white arrow: nephric duct; *: urogenital sinus. Scale bars: 100 µm.

Supplemental Figure 2. Cre activity in *Isl1Cre* mice. Sections of *Isl1Cre;tdTomato* reporter mice at E14.5 were stained with an anti-RFP antibody. The signals are observed in the metanephric and ureteral mesenchyme (A), ureteric mesenchyme at the distal ureter (B), urinary bladder (C), and genital tubercle (D). No Cre activity is detected in the ureteric epithelia. Scale bars: 100 μm.