



**Figure 6–Figure Supplement 1. Skeletal alterations in *Pcgf6*<sup>KI/KI</sup> and *Pcgf6*/*Pcgf2* dKO newborn mice.**

(A) *Pcgf6* expression was high in ESCs and in testis, among different cells and tissues. Data was retrieved from BioGPS (biogps.org). (B) Analysis of *Pcgf6* expression during early embryogenesis (GEO dataset # GSE66582); and during differentiation from ESCs to EpiLCS, and then to PGCLCs (GEO dataset # GSE86903). Gene expression was measured in RPKM. (C) Schematic representation of the construct for conditional expression of 3xFlag-tagged *Pcgf6* cDNA from the *Rosa26* locus. The targeting construct contains a 3xFlag-tagged mouse *Pcgf6* cDNA downstream of an loxP (closed triangles)-flanked neomycin resistance gene (neo). (D) Skeletal alterations in *Pcgf6*<sup>+/+</sup> and *Pcgf6*<sup>KI/KI</sup> newborn mice. (a,b) Lateral views of the cervical and thoracic regions revealed the prominent spinous process on 8 in *Pcgf6*<sup>KI/KI</sup> (b, indicated by an arrow). (c, d) Ventral views of the rib cages identified ectopic sternal ribs on 7 in *Pcgf6*<sup>KI/KI</sup> (d, labeled C7). (E) Skeletal alterations in *Pcgf6*<sup>+/+</sup>; *Pcgf2*<sup>-/-</sup> and *Pcgf6*<sup>-/-</sup>; *Pcgf2*<sup>-/-</sup> newborn mice. (a, b) Lateral views of the cervical and thoracic regions. An ectopic rib on 7 in *Pcgf2*<sup>-/-</sup> was not observed on *Pcgf6*<sup>-/-</sup> background (a, indicated by an arrowhead). (c, d) Ventral views of the rib cages from the specimens a and b, respectively, showing the ectopic rib (c, indicated by arrowheads). The posteriormost sternal ribs of *Pcgf2*<sup>-/-</sup> were on 13 (c, labeled r6) but on 14 on *Pcgf6*<sup>-/-</sup> background (d, labeled r7). (e, f) Ventral views of the thoracic and lumbar regions. The posteriormost ribs of *Pcgf2*<sup>-/-</sup> were on 19 (e, labeled T12) but on 20 on *Pcgf6*<sup>-/-</sup> background (f, labeled T13). (F) Schematic representation summarizing the axial alterations in *Pcgf6*<sup>-/-</sup>, *Pcgf6*<sup>+/+</sup>, *Pcgf6*<sup>KI/KI</sup>, *Pcgf6*<sup>+/+</sup>; *Pcgf2*<sup>-/-</sup> and *Pcgf6*<sup>-/-</sup>; *Pcgf2*<sup>-/-</sup> newborn mice. Each arrow represents the following morphological changes in the vertebrae; (a)–(e) See Figure 6; (f) 7→8: ectopic rib(s) on 7; (g) 8→9: the prominent spinous process on 8; (h) 14→15: ribs on 14 not fused to the sternum; (i) 20→21: loss of ribs on 20.