

Figure S1. mTOR is critical for embryonic skeletal development. (A-B) Gross appearance of E18.5 wild-type (A) versus mTORCKO (B) littermates. (C-D) Whole-mount skeletal staining of E18.5 WT (C) and mTORCKO (D) littermates. (E-L) Higher magnification of calvaria (E,F), and sterna (G,H) from E18.5 WT (E, G) and mTORCKO (F, H) littermates. (I-J) H&E staining of longitudinal sections through the ulna from E18.5 WT (I) and mTORCKO (J) littermates. Green arrows: parietal bone; red arrows: sternum. BM: bone marrow; *: hypertrophic cartilage.

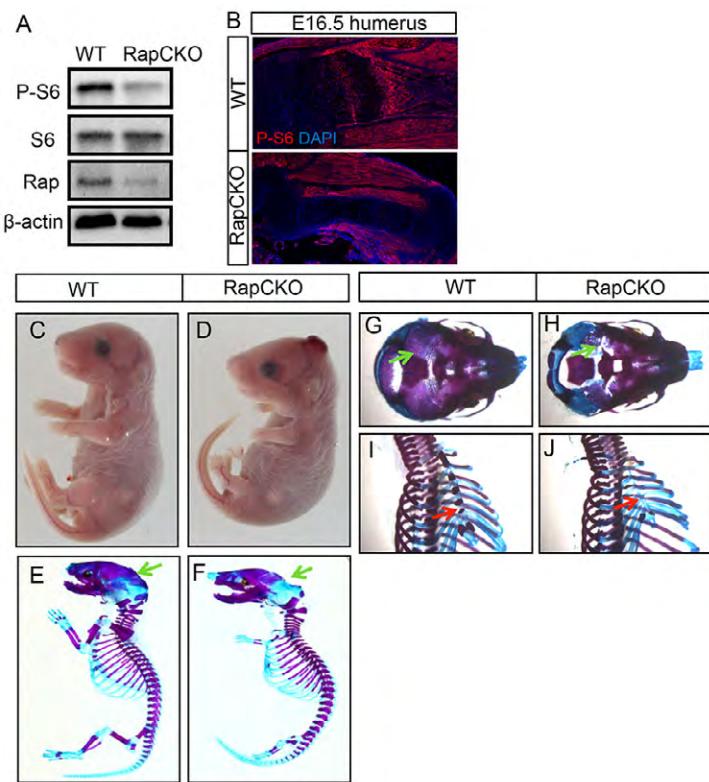


Figure S2. Raptor critically regulates embryonic skeletal development. (A) Western blot analyses with forelimb buds from E12.5 WT versus RapCKO littermates. (B) P-S6 immunofluorescence staining on longitudinal humeral sections from E16.5 WT versus RapCKO embryos. P-S6: red; DAPI: blue. (C-D) Gross appearance of E18.5 wild-type (C) versus RapCKO (D) littermates. (E-F) Whole-mount skeletal staining of E18.5 WT (E) and RapCKO (F) littermates. (G-J) Higher magnification of calvaria (G, H), and sterna (I, J) from E18.5 WT (G, I) and RaptorCKO littermates (H, J). Green arrows: parietal bone; red arrows: sternum.

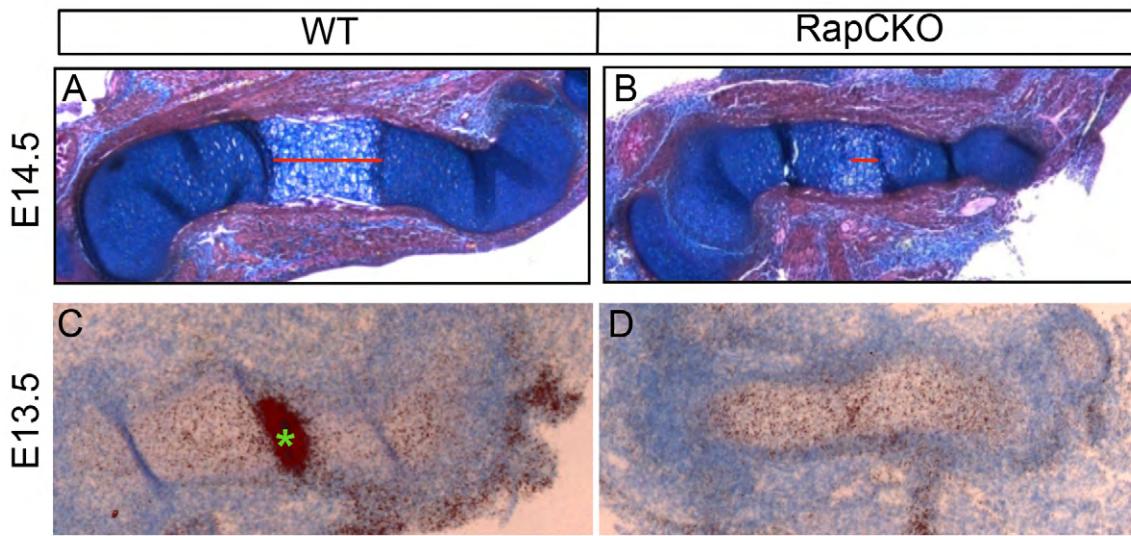


Figure S3. mTORC1 regulates chondrocyte hypertrophy. (A, B) Alcian Blue/Picosirius Red staining of longitudinal humeral sections from E14.5 WT versus RapCKO littermates. Red lines indicate the length of hypertrophic zone. (C, D) In situ hybridization of *Col10a1* on longitudinal humeral sections from E13.5 littermates. Hybridization signal in red. Asterisk denotes *Col10a1*-expressing domain in WT sample.

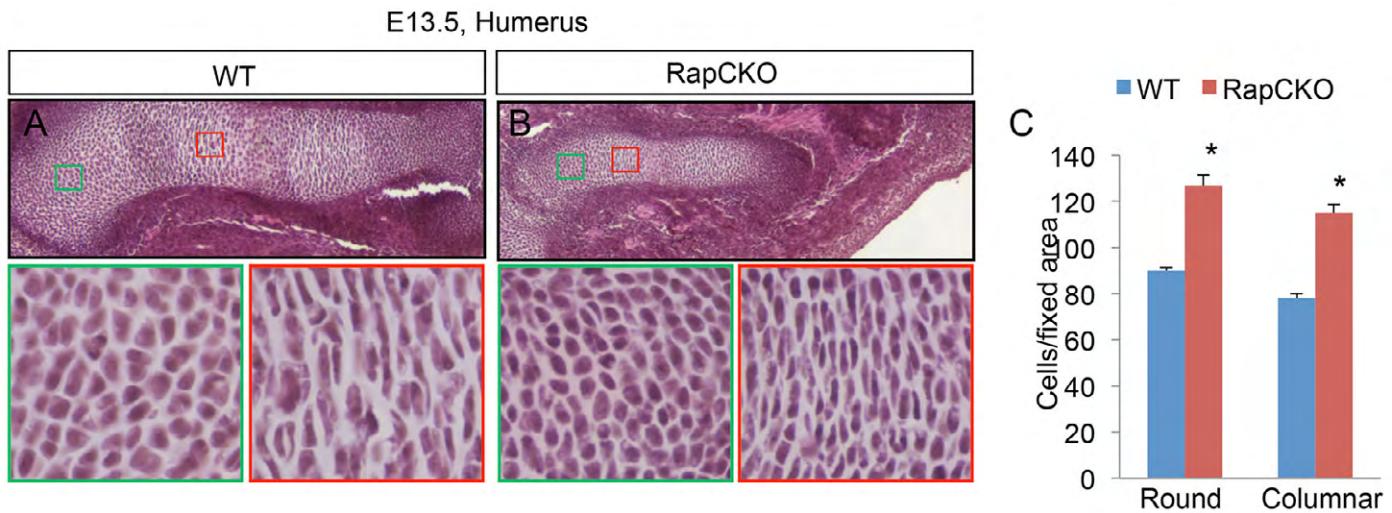


Figure S4. mTORC1 regulates chondrocyte size and amount of extracellular matrix. (A-B) H&E staining of longitudinal humeral sections from E13.5 WT (A) versus RapCKO (B) littermates. Areas in colored boxes shown below at higher magnification. (C) Quantification of chondrocyte density in round and columnar regions. *: p<0.05, n=3.