

Fig. S1. Appendicular and axial skeletal development is normal in *Ptprs*^{-/-}; *Ptprf*^{-/-} embryos. Alizarin Red and Alcian Blue stainings of bone and cartilage. (**A,B**) Bone and cartilage development of the DKO body is normal at E18.5. (**C-F**) Bone and cartilage development of the DKO body (C,D) and middle ear capsule (E,F) are normal at E14.5. Note that Meckel's cartilage articulates normally with the middle ear. MC, Meckel's cartilage; ME, middle ear.

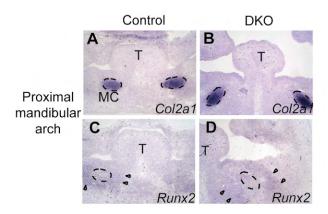


Fig. S2. Proximal Meckel's cartilage is specified normally in *Ptprs*^{-/-}; *Ptprf*^{-/-} **embryos.** (**A-D**) *In situ* hybridization using probes to *Col2a1* and *Runx2* reveals normal proximal Meckel's cartilage specification in DKO mandibular arch at E12.5. Differentiation of proximal bone precursors also occurs normally (arrowheads). MC, Meckel's cartilage; T, tongue.

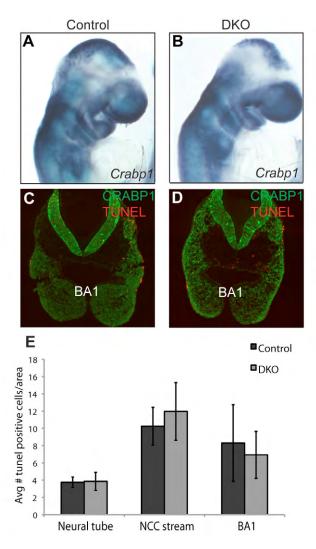


Fig. S3. Neural crest migration and survival in the first branchial arch. (A,B) Whole-mount *in situ* hybridization using a probe for cellular retinoic acid binding protein (CRABP1) as a marker of migratory neural crest cells reveals normal migration in DKO embryos at E9.5. (C-E) Immunofluorescent labeling of CRABP1 and TUNEL reveals no significant difference in neural crest cell (NCC) apoptosis in either the migratory stream or first branchial arch (BA1) at E9.5.

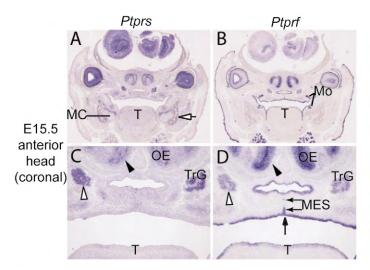


Fig. S4. Expression of *Ptprf* and *Ptprs* during palatal shelf fusion. *In situ* hybridization using probes for *Ptprf* (A,C) and *Ptprs* (B,D) on coronal E15.5 sections. (A,C) *Ptprf* is expressed in the medial epithelial seam (MES, black arrows) where palatal shelf fusion occurs, as well as throughout the oral epithelium. (B,D) *Ptprs* is expressed diffusely in the palatal shelf mesenchyme and in ossification centers surrounding Meckel's cartilage (open arrow). Additional regions of strong LAR family expression are indicated by arrowheads. MC, Meckel's cartilage; Mo, molar tooth buds; OE, olfactory epithelium; T, tongue; TrG, trigeminal ganglion.

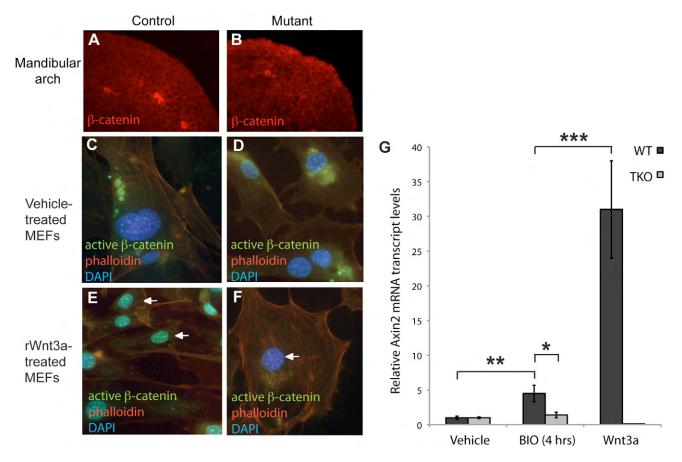
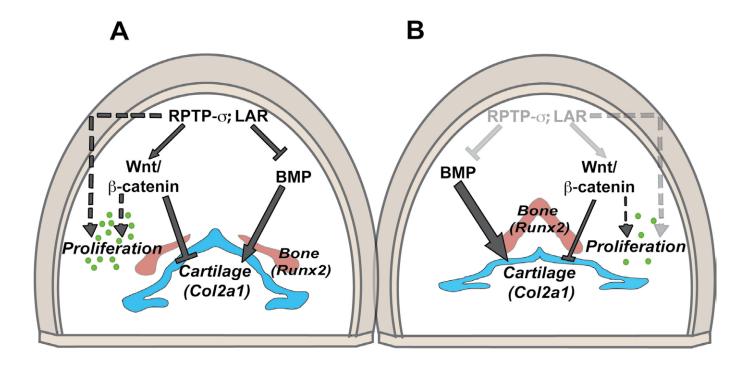


Fig. S5. β-catenin activation is impaired in mouse embryonic fibroblasts mutant for LAR family phosphatase activity. (A,B) Immunofluorescent staining for total β-catenin protein on coronal E10.5 pharyngeal arch sections. There is no significant difference in β-catenin levels between control (A) and $Ptprs^{-/-};Ptprf^{-/-}$ (B) embryos. (C-F) Immunofluorescent staining of (C,E) wild-type (WT) mouse embryonic fibroblasts (MEFs) and (D,F) $Ptprs^{-/-};Ptprf^{-/-};Ptprd^{-/-}$ triple knockout (TKO) MEFs for Ser27/37 dephosphorylated (active) β-catenin reveals a specific defect in the activation of β-catenin in TKO MEFs upon rWNT3A stimulation (arrows). (G) qPCR analysis of Axin2 transcription reveals that treatment with the GSK3β inhibitor BIO alone is insufficient to restore β-catenin activity, confirming the specificity of the rescue to the Wnt pathway. *P<0.1, **P<0.05, ***P<0.005.



Wild type

Double knockout

Fig. S6. Model for LAR family phosphatase activity during pharyngeal arch development. (**A**) Normally, PTPRS (RPTPσ) and PTPRF (LAR) act to dampen Bmp signaling activity and promote canonical β-catenin activity, which serves to correctly specify Meckel's cartilage and downstream initiation of mandibular bone development. In addition, PTPRS and PTPRF promote mandibular arch proliferation, potentially through their action on canonical Wnt signaling. (**B**) In *Ptprs*^{-/-}; *Ptprf*^{-/-} embryos, loss of the phosphatases permits concomitant intensification of Bmp signaling and downregulation of canonical Wnt restriction of cartilage development, thereby altering the specification of Meckel's cartilage and indirectly affecting mandibular bone positioning. Loss of the LAR family also disrupts proliferation, further affecting mandibular development.

Table S1. Primer sequences for qPCR or ISH

Gene	Use	Forward primer	Reverse primer
beta-microglobin	qPCR	GACTGGTCTTTCTATATCCTGG	CTTTCTGCGTGCATAAATTG
Axin2	qPCR	CCATTTTGGACGACCACCTCTC	GAAGAAGGGTATGACACTGCTGATG
cyclin D1	qPCR	TCTTTCATTGGGCAACGGG	TCCTCAGTTTGGATGGCTCTCC
Ptc1	qPCR	TCTGCTGGGTGTACTGATGC	TCAGGACACGGTCCAAAGA
Msx1	qPCR	TCCTCAAGCTGCCAGAAGAT	TTGGTCTTGTGCTTGCGTAG
DIx1	qPCR	TTCATCTGACGCTGAGTGTTGG	TTTCCTGTCCTTGTTCCCTCTTC
DIx5	qPCR	TCAGGAATCGCCAACTTTGC	TGCCATAAGAAGCAGAGGTAGGAG
Gsc	qPCR	TAAGAACCGCCGAGCCAAG	CCGAGTCCAAATCGCTTTTACC
Sox9	qPCR	TCGGAACTGCCTGGAAACTTC	GAGGGAGGAAAACAGAGAACG
Col2a1	qPCR	GAGCAGCAAGAGCAAGGAAAAG	CAGTGGACAGTAGACGGAGGAAAG
Runx2	qPCR	ACCAGTCTTACCCCTCCTATCTGAG	GCAGTGTCATCATCTGAAATACGC
Msx1	ISH	CCTACGCAAGCACAAGACCAAC	TGGGGACCACGGATAAATCTC
Col2a1	ISH	CATTATTGACATCGCACCC	CAGAATAGCACCATTGTGTAGG
Runx2	ISH	GAGCTATTAAAGTGACAGTGGACGG	CAACACCATCATTCTGGTTAGGC
Crabp1	ISH	CAGCAGAGGTGGGTGCCTGCCC	GCAGCCAACCAGTTTAATGACTGG

All primers are shown 5' to 3'.