

Fig. S1. β -catenin is required for cranial bone fate in paraxial mesoderm osteoprogenitor cells. X-Gal (A-C) and AP (D,E) staining or in situ hybridization (F-K) on tissue sections. Dashed lines, skull progenitors (A,B,F) or *En1Cre* lineage (H-K). White arrows indicate absent *Lef1* expression (G). Black arrows point to ectopic *Sox9* expression (K). Inset (B) shows plane of section. Tel, telencephalon; dm, dura mater; se, surface ectoderm; pp, parietal bone progenitor cells. Scale bar: 100 μ m.

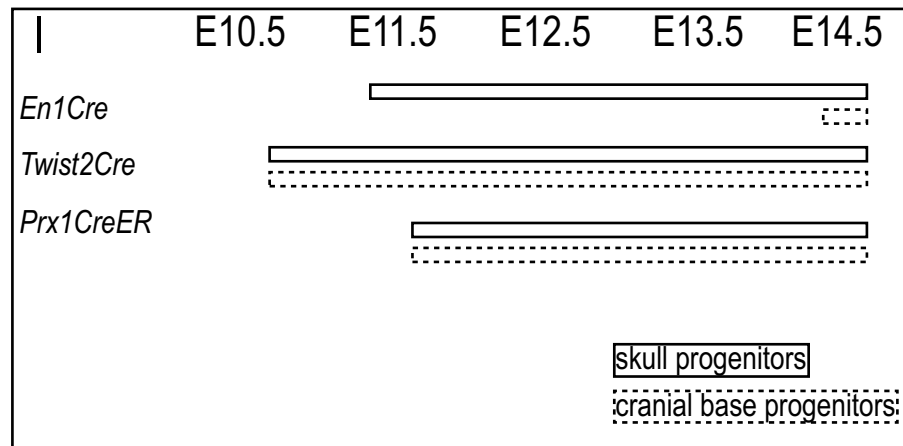
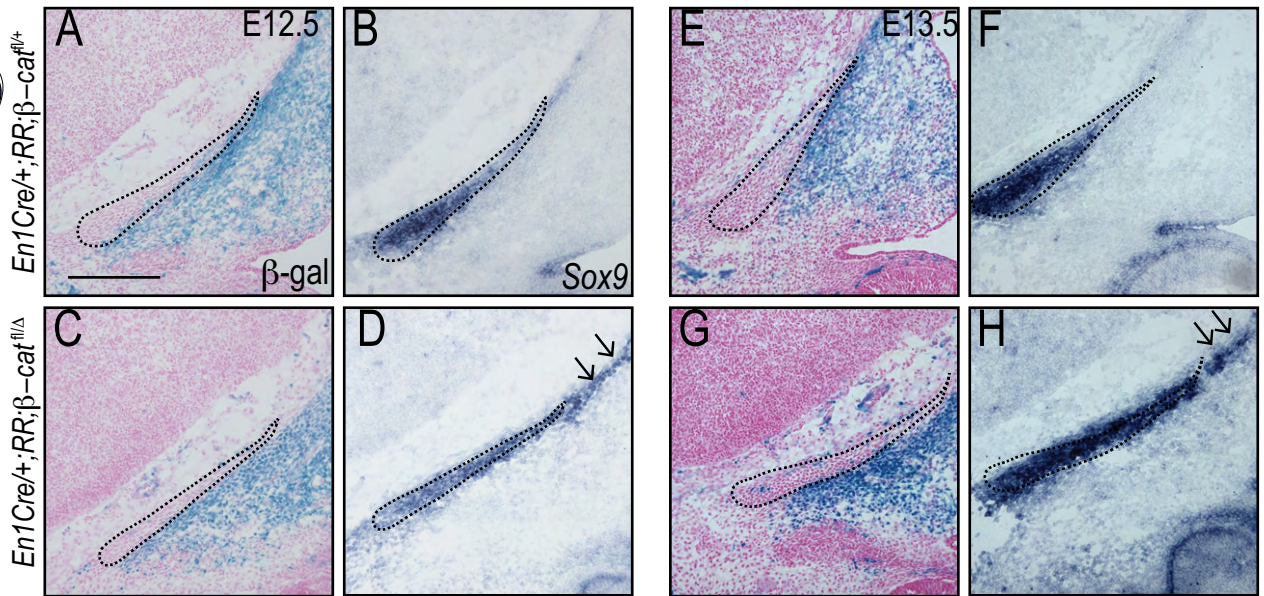
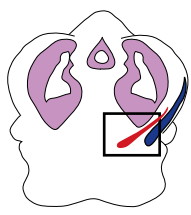


Fig. S2. Lack of contribution of *En1Cre* lineage to skull base and description of temporal differences in Cre activity. (A-H) X-Gal staining (A,C,E,G) or in situ hybridization (B,D,F,H) on coronal sections. Arrows (D,H) indicate ectopic *Sox9* expression. (I) Onset of Cre recombinase activity for the lines used in this study. Scale bar: 100 μ m.

Trigeminal Ganglion

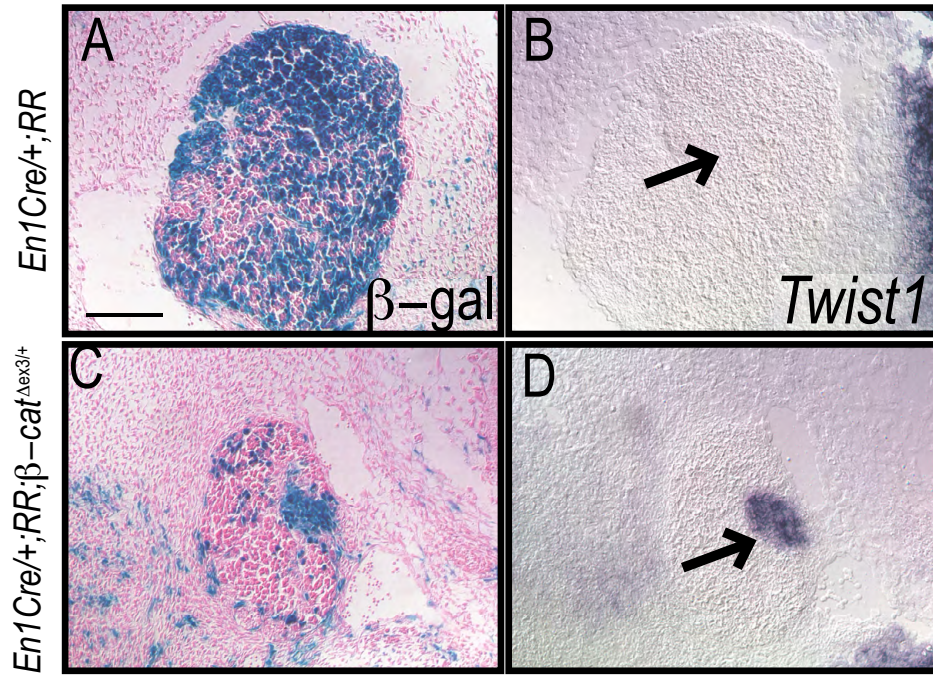


Fig. S3. β -catenin is sufficient for ectopic *Twist1* expression. Coronal tissue sections through trigeminal ganglia stained with X-Gal (A,C) or hybridized with mRNA probes (B,D). Arrows indicate area of ectopic expression. Scale bar: 100 μ m.

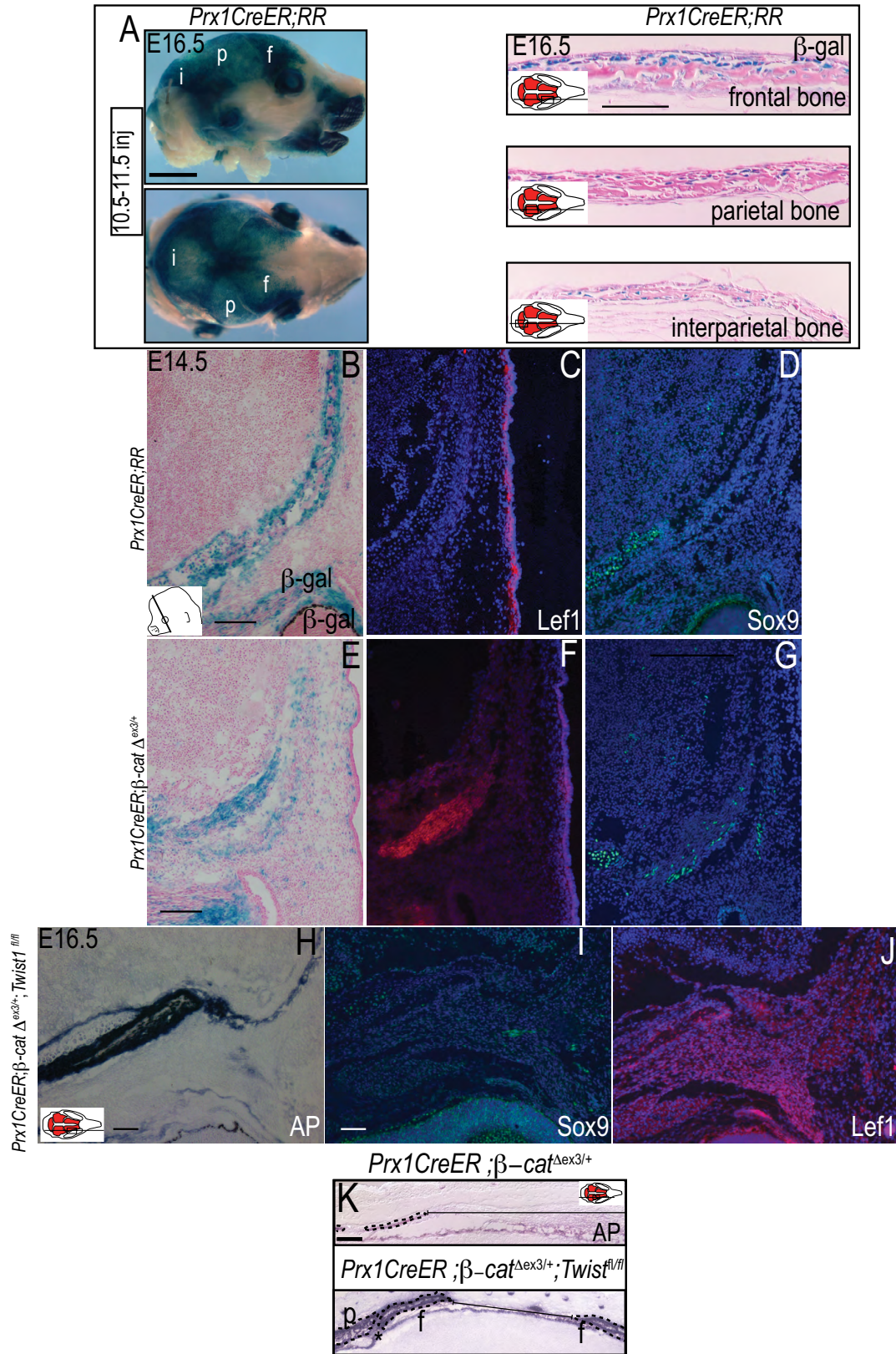


Fig. S4. Activation of β -catenin results in Lef1 expression; deletion of *Twist1* restores AP in Lef1-expressing cells. (A,B,E) Whole-mount X-Gal staining performed with skin removed and sagittal (A) or coronal (B,E) sections counterstained with Eosin. (C,D,F,G,I,J) Immunofluorescence on sections. (H,K) AP staining on sections. (K) Black brackets indicate missing bone, black dashed lines indicate AP⁺ bone, and asterisk indicates the coronal suture. Scale bars: 100 μ m for tissue sections; 25 mm for whole-mounts.

Table S1. Primer sequences used for ChIP-qPCR

H3K4me1, β-catenin ChIP-Twist1 qPCR			
Twist1 -62	E1	ATGGGTGGCAGAGGTGATTA	CATGGGCTCCCATTTATAGC
Twist1 -60	E2	TTAGCCAGAGATCCCCGATT	CCTTGTGCCATTTCATCATCA
Twist1 -22	E3	CCTTCTCCCCATCATTGAAA	ACCATGCCAGCCTTTTGTAT
Twist1 -18	E4	TGGACTTTGCCGAAAAGAAA	ACCTCCCGGTTCTTATTGAG
Twist1 -4.5	E5	CTCTTGTGGTCACTGGCAAA	TCCTACTCTCAGCCCTCCAA
Twist1 -3	E6	GAAAACAAAGCCCCTTGTGA	AGGCTTCTGTCAATCTGGTGA
Twist1 +5	E7	AACCCCCGCTTAATGTTCTT	ACCTGCCATAGGCTCCTTTT
Twist1 +10	E8	CTACACCCCAACCCTTGGT	GACTACTCCACCCCTCAAATC
Twist1 +16	E9	TGAAATGTAACCCTGGTCCAA	TCAGCATTCAAGAGCCAAAA
Twist1 -1.7	P1-A	GGCACACTGTGGTCATTGTC	AAACTCTGGGGTGTGGAATG
Twist1 -1.8	P1-B	GGGTTTACTGGCCTCAGAAGA	CTGGGTCTGCTGAAGCTCT
Twist1 -1.9	P1-C	CTTTGGTCCAACCCAGAAAG	GCTGAGGACCTTGCTAAACG
Twist1 -75	NT1	TGTGATAAGGCACCACCTCA	GCCCTGCTCTTCAAACCTCAC
Twist1 -37	NT2	CAGGACAAGAGGAAGCAACC	ATGCACAACAGCACCACAAT
Twist1 -26	NT3	TCTGCAATTAGCCTTCTTTCC	AGCTAACAGCCCTGGTTTTG
Twist1 -8	NT4	CTGCACAATGCTGGTGTCT	TGGCAAATGGAGAATGATGA

Twist ChIP-Sox9 qPCR		
	F:	R:
3'UTR 1	AAGCACATTTTCCCTGGTTG	AAAAATGAAAACCTTCTCCTTACAAAA
3'UTR 2	TTTTTCACGCAGCCCTAAGT	AAAAATGAAAACCTTCTCCTTACAAAA
3'UTR 4	CAAGCACATTTTCCCTGGTT	CGCTGGTATTCAGGGAGGTA
NT1	GGTTTTGAAACGATGTGCAG	TGTGTATCGGAGCCACCTG
NT2	TAGCCGACAAGGTTGGTTTT	ACTGTGTATCGGAGCCACCT
NT4	CGCTGTAGCACAAGGACAGA	TCAGCTAGCGTAGCCTCCTC
Promoter1	AGCGCCTCTGCTAAGTGC	ATGAAGGGGTCCAGGAGATT
Promoter2	CCGGTTTCGTTCTCTGTTTT	GCGGGCACTTAGCAGAGG
Promoter3	GGGAGCGACAACCTTACCAG	AGGAGGGAGGGAAAACAGAG