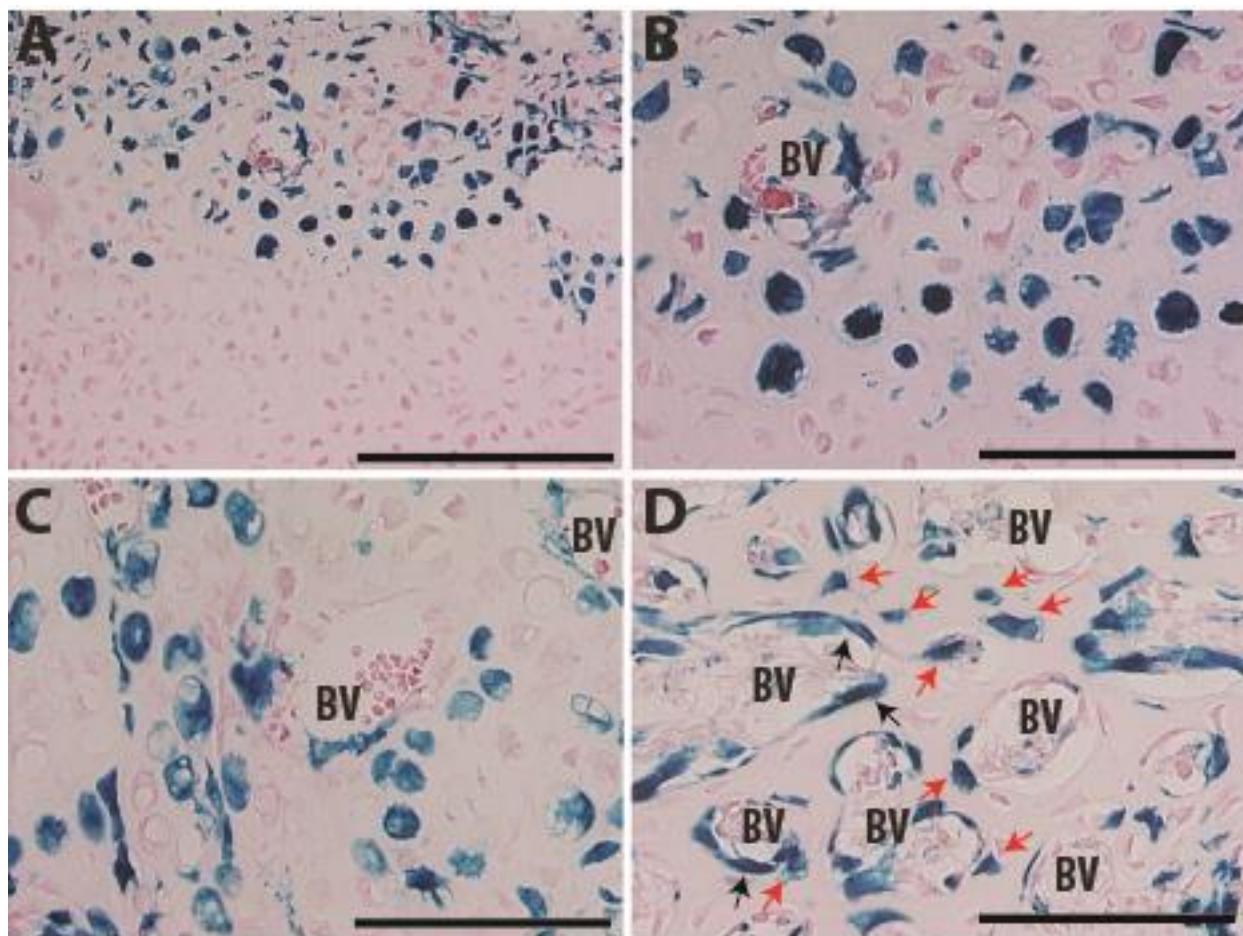
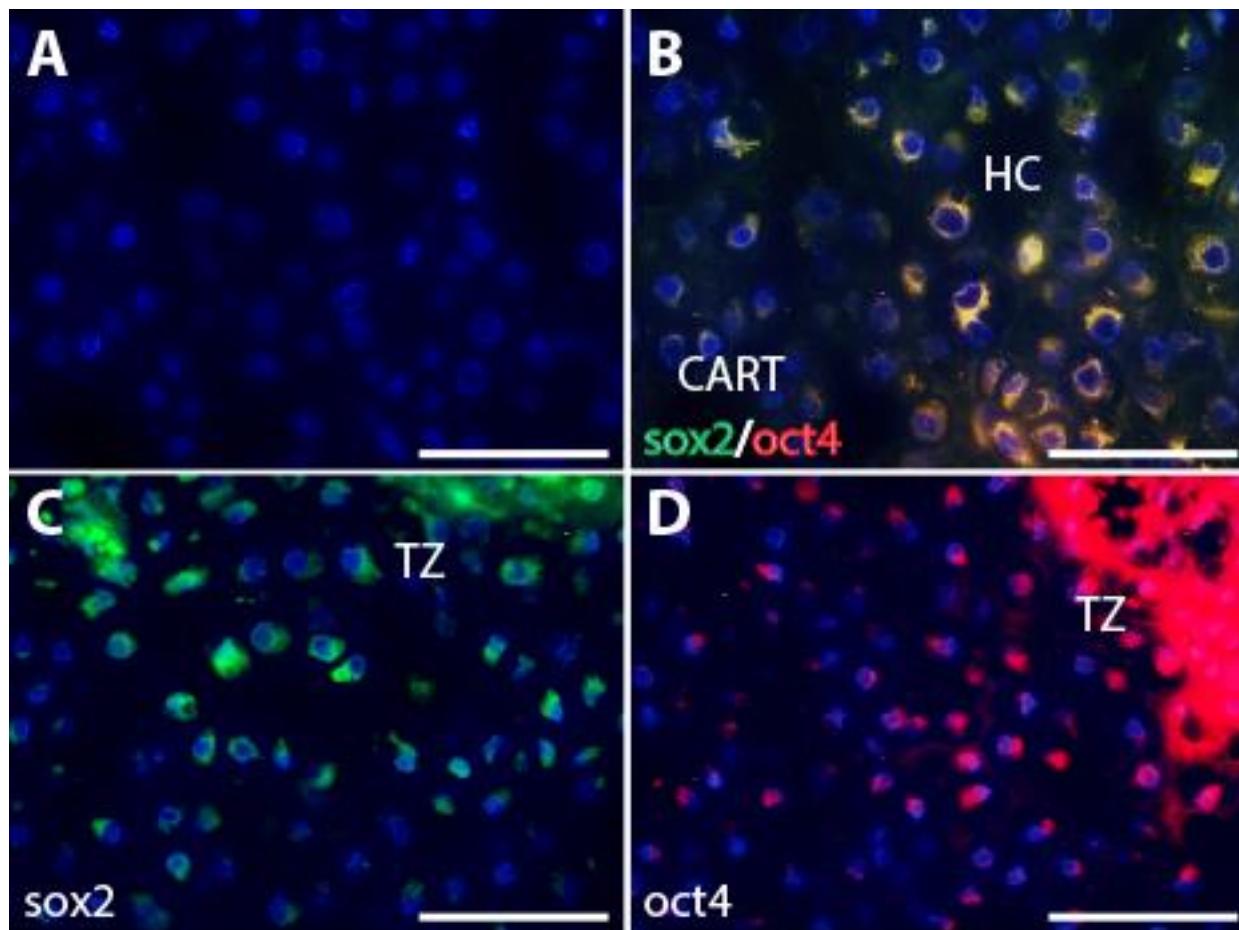


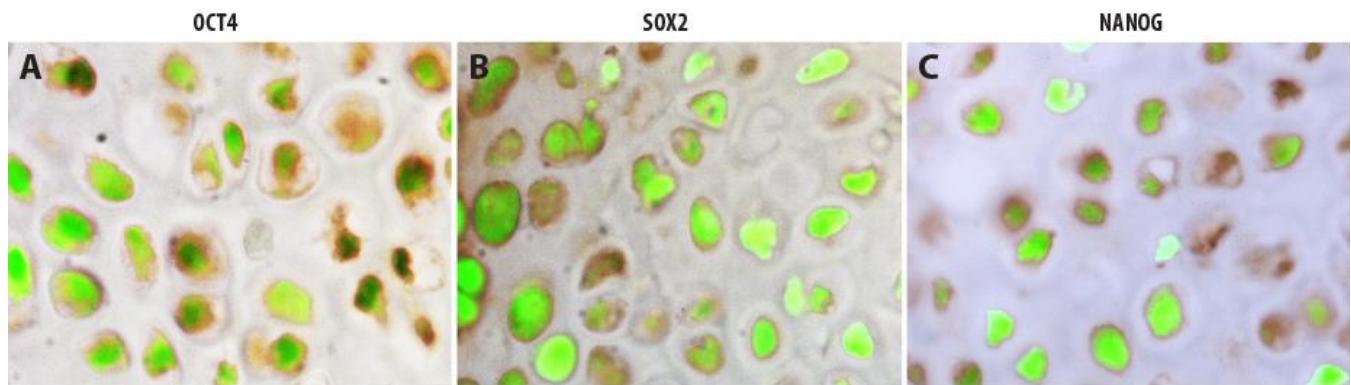
**Figure S1: Osteopontin expression at transition zone in the fracture callus.** *In situ* hybridization for *Opn* expression in the day-10 post-injury fracture callus. (A) Low magnification of the fracture callus, (B) *Opn*-expressing hypertrophic chondrocytes away from the TZ, (B) *Opn*-expressing hypertrophic chondrocytes in TZ, (D) *Opn*-expressing cells in the bone matrix. CART = cartilage NB = newly formed bone, HC = hypertrophic chondrocytes, (A) Scale = 500μm, (B-D) Scale = 100μm



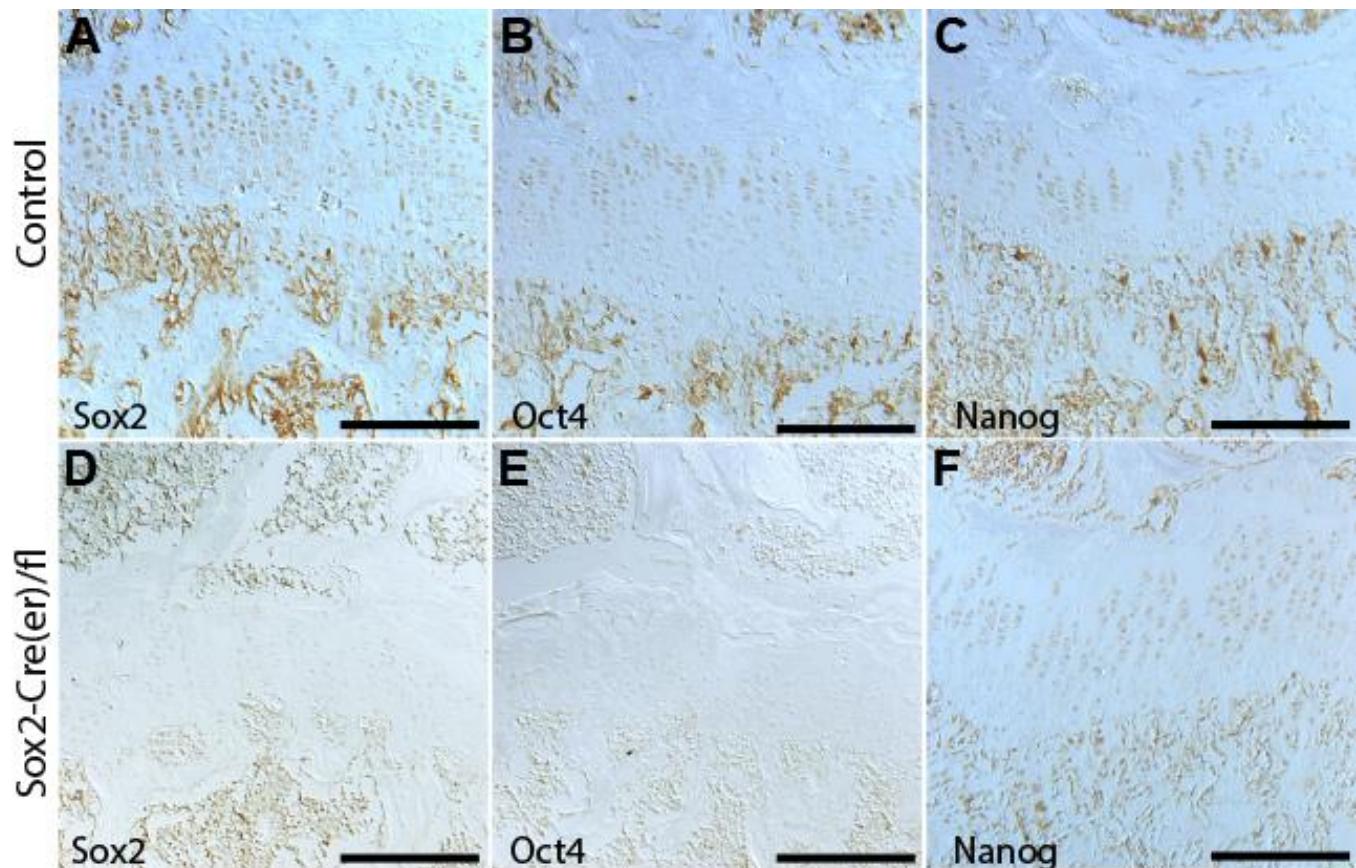
**Figure S2: *Osterix* expression in the transition zone of the fracture callus.** The *Osterix*(Sp7)-CreERT::R26R mouse was generated, fractured, and tamoxifen administered from days 6-10 post-injury. The fractures were collected at 14-days post-injury and x-gal staining performed on frozen sections to identify  $\beta$ -galactosidase-expressing cells (blue) in the TZ of the fracture callus. (A) Low magnification of the TZ, (B-C) high magnification of *Osterix*-expressing hypertrophic chondrocytes in TZ, (D) *Osterix*-expressing osteoblast/cytes and bone lining cells in the TZ. (A) Scale = 200 $\mu$ m, (B-D) Scale = 100 $\mu$ m



**Figure S3: Co-expression of Sox2 and Oct4 at transition zone in the fracture callus.** Using frozen sections from a day 14 wild type murine fracture we used different SOX2 (#371R-17) and OCT4 (#309M-17) antibodies from Cell Marque Corporation to confirm IHC expression in the main manuscript. (A) No expression of SOX2 or OCT4 is seen in immature chondrocytes, compared to (B) co-localization and (C-D) to robust expression in hypertrophic chondrocytes at the TZ. Scale = 100 $\mu$ m.



**Figure S4: Cells from the chondrocyte lineage express pluripotent markers.** (A-C) Cells from the chondrocyte lineage were labeled in *Agc1CreER<sup>T</sup>::Ai9* mice (pseudo-colored green) via tamoxifen injections as described in main manuscript and co-stained for (A) OCT4 (B) SOX2, (C) NANOG using immunohistochemistry (brown).



**Figure S5: Pluripotency Markers in the Adult Murine Growth Plate.** Immunohistochemistry on growth plate in adult (A-C) C57B6 mouse versus (E-F) *Sox2Cre<sup>ERT/fl</sup>* mouse: immunohistochemistry (A,D) SOX2 (B,E) OCT4, (C,F) NANOG. Scale = 200μm.

**Table S1. Mouse database**

Type	Abbreviated Name	Strain	Jackson #
Wild Type	B6	C57Bl/6J	000664
Reporter	R26R	Gt(ROSA)26Sor <sup>tm1Sor</sup>	003309
	Ai9	B6.Cg-Ct(ROSA)26Sor <sup>tm9(CAG-tdTomato)Hze/J</sup>	007909
	ROSA <sup>mT/mG</sup>	B6.129(Cg)-Gt(ROSA)26Sor <sup>tm4(ACTB-tdTomato,-EGFP)Luo</sup>	007676
CreER <sup>T</sup>	Col2-CreER <sup>T</sup>	FVB-Tg(Col2a1-cre/ERT)KA3Smac/J	006774
	Agc1-CreER <sup>T</sup>	Agc1 <sup>tm(IRES-CreERT2)</sup>	019148
	Oct4-CreER <sup>T</sup>	B6(SJL)-Pou5f1 <sup>tm1.1(cre/Esr1*)Yseg/J</sup>	016829
	Sox2-CreER <sup>T</sup>	B6;129S-Sox2tm1(cre/ERT2)Hoch	017593
	Osx-CreER <sup>T</sup>	B6.Cg-Tg(Sp7-tTA,teO-EGFP/cre)1Acc/J	006361
CKO	Sox2 <sup>CreERT/fl</sup>	Sox2-Cre-ERT2(Ki)Sox2::LoxP(Ki)R26-mTmG(Ki/+)	

**Table S2. Antibody database**

Antibody	Company	Cat. no.	Dilution
β-catenin	Abcam	ab32572	1:500
Caspase-3	Cell Signaling Technology	9661	1:1600
Oct4	Abcam	ab19857	1:200
Osteocalcin	Abcam	ab93876	1:200
Nanog	Abcam	ab80892	1:200
PECAM/CD31	BD Biosciences	557355	1:100
Runx2	Abcam	ab23981	1:200
Sox2	Abcam	ab97959	1:200
Sox9	Abcam	ab26414	1:200
VEGF	Abcam	ab46154	1:200

**Table S3. Primer list**

SYBR Green Primer Sequences			
	Forward (5' to 3')	Reverse (3' to 5')	
GAPDH	TGATGACATCAAGAAGGTGGTGAAG	CCTTGGAGGCCATGTAGGCCAT	
Sox2	GGCAGCTACAGCATGATGCAGGAGC	CTGGTCATGGAGTTGACTGCAGG	
Oct4	CTGTAGGGAGGGCTTCGGGCACTT	CTGAGGGCCAGGCAGGAGCACGAG	
Nanog	AACCAGTGGTTGAATACTAGCAATG	CTGCAATGGATGCTGGGATACT	
Col2a1	GGCTCCCAGAACATCACCTA	TCGGCCCTCATCTCTACATC	
Col10a1	AATCTGAAATGCAAGGTGCT	AAGACTCAAATAGTCATTAAAGCAAA	
Col1a1	CCCAGAACATCACCTATCAC	TTGGTCACGTTCAAGTGGTC	
Osteocalcin	CGCTCTGTCTCTGACCTC	TCACAAGCAGGGTTAAGCTC	