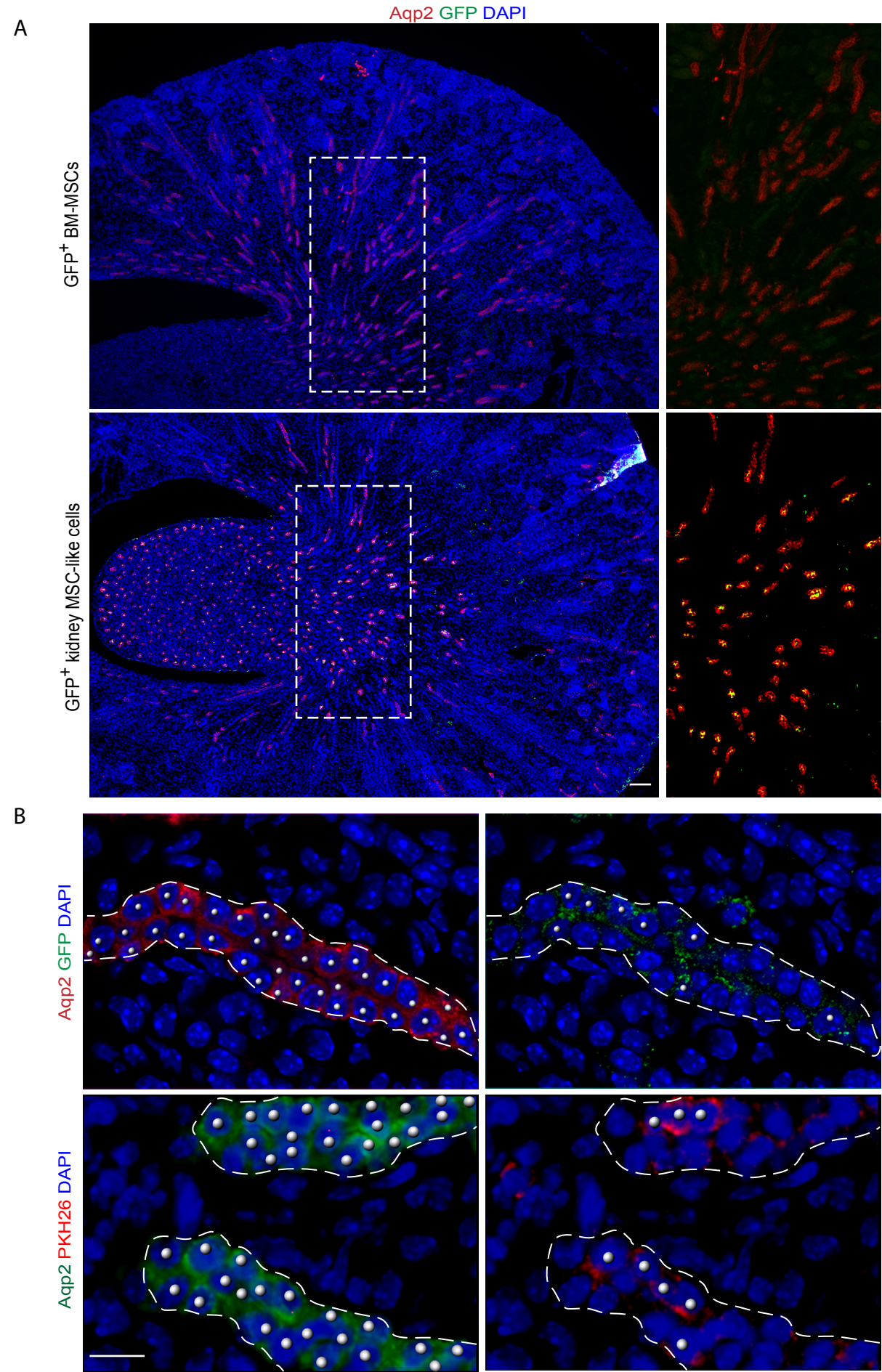
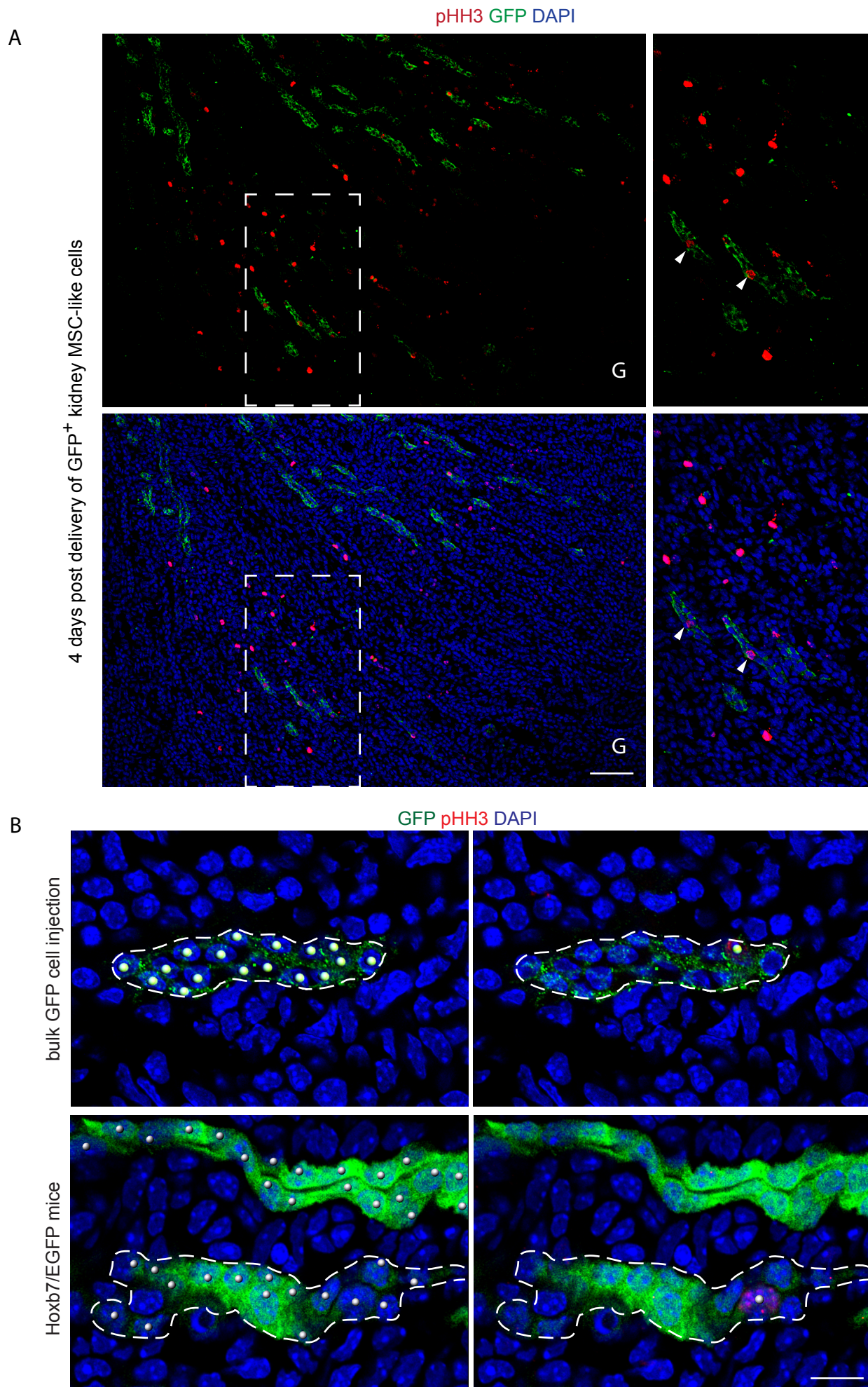


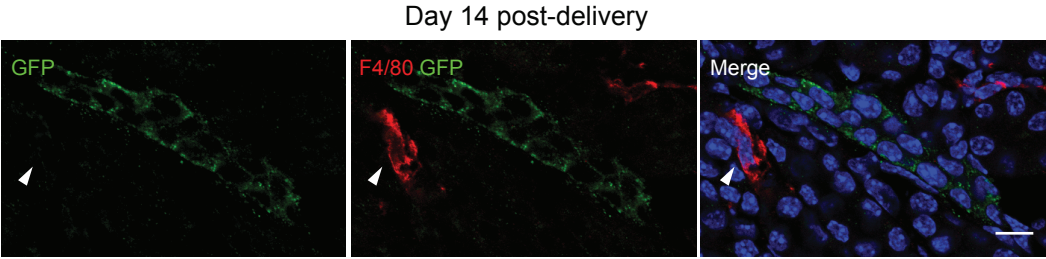
Supplementary Figure 1



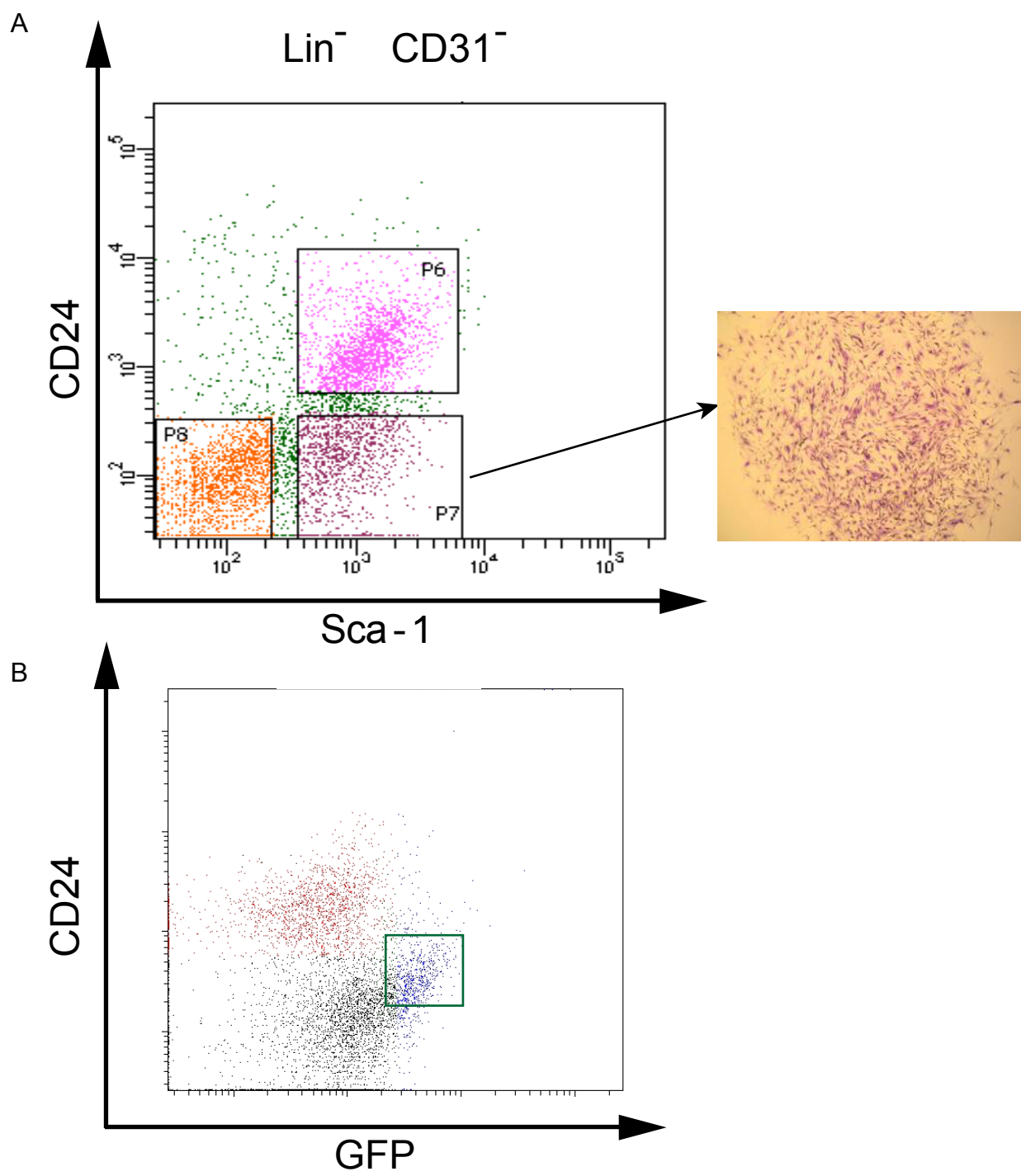
Supplementary Figure 2



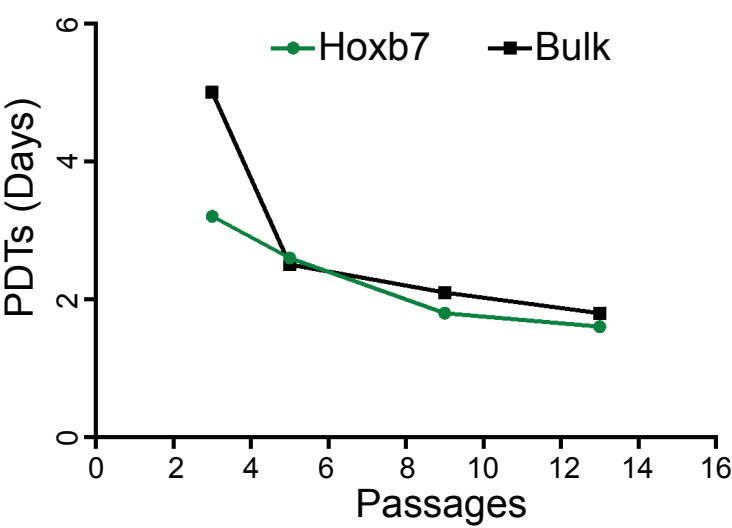
Supplementary Figure 3



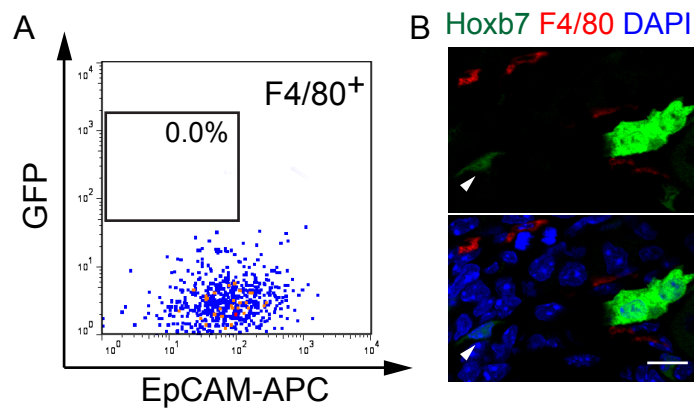
Supplementary Figure 4



Supplementary Figure 5

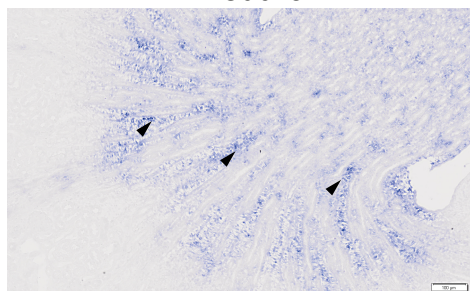


Supplementary Figure 6

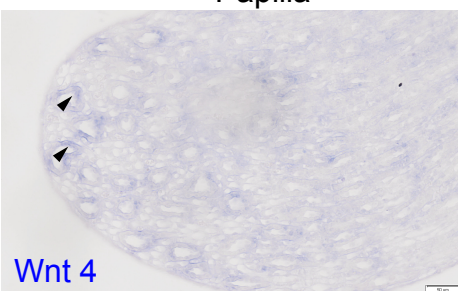


Supplementary Figure 7
Medulla

PND 6

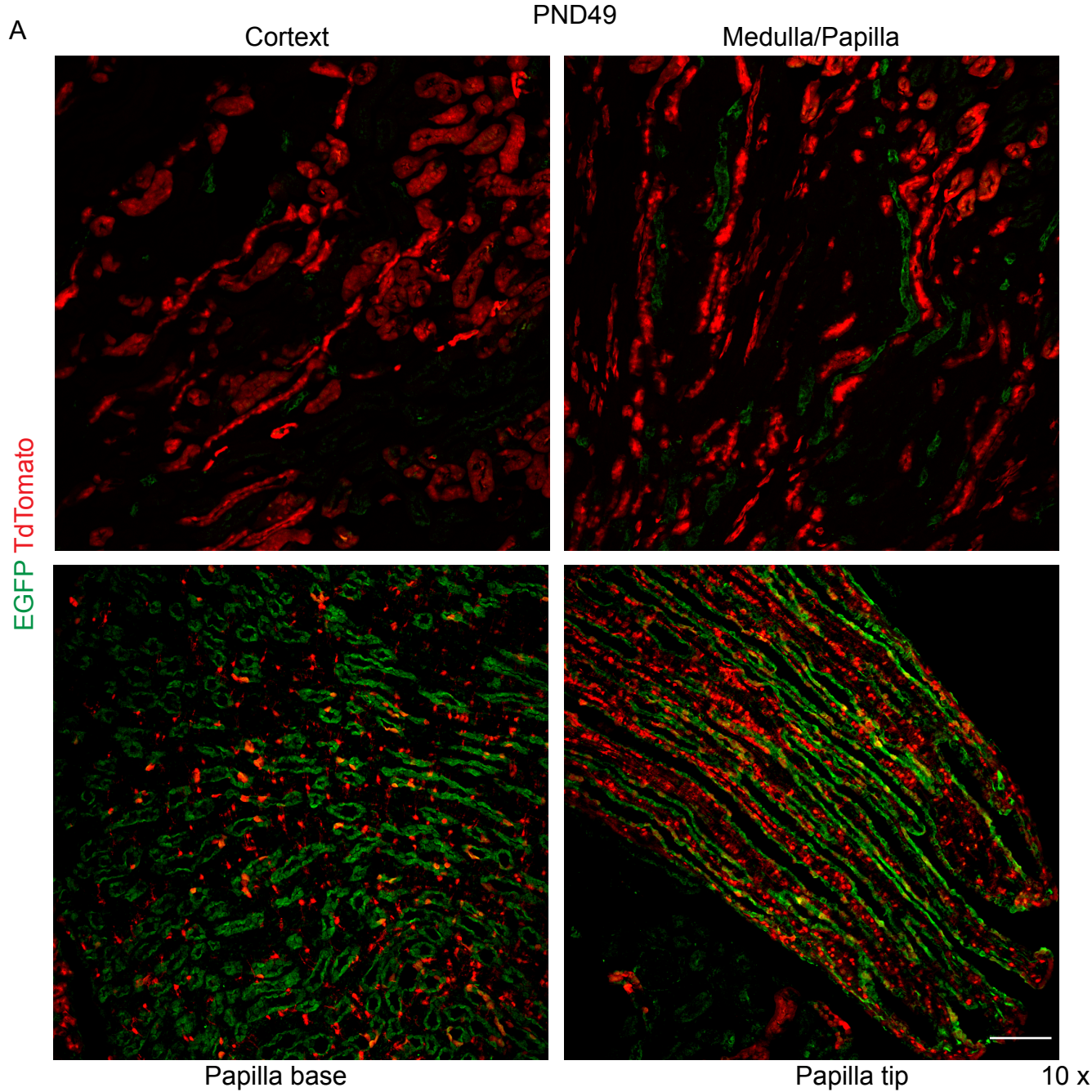


Papilla



Wnt 4

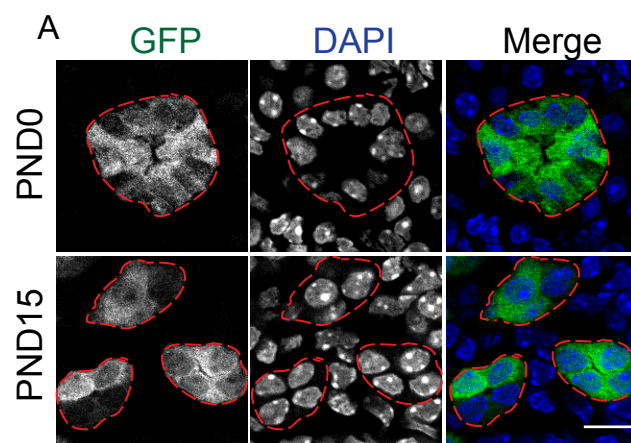
Supplementary Figure 8



B

	Collecting duct Aqp2 ⁺ DAPI ⁺				Wnt4-expressing EGFP ⁺ DAPI ⁺		
	EGFP ⁺	TdTomato ⁺	double	total	Aqp2 ⁻	TdTomato ⁺	total
E19.5	0	0	0	813	371	31	371
PND49	878	73	73	878	0	73	878

Supplementary Figure 9



Supplementary Table 1

qPCR primer sequences

Gene	Forward Primer (5' – 3')	Reverse Primer (5' – 3')
GAPDH	CCCAATGTGTCCGTCGTG	GCCTGCTTCACCACCTTCT
NG2	CCCGATGATGTAGGTGATGCTTC	GCTGCCCTGTAGTGAAACACAAC
Pod1	CCCACTAAGAAAAGCCCGCTC	CCGTTCTCGTACTTGTCGTTG
Wnt4	AGTGGAGAACTGGAGAAGTGT	CAAAGGACTGTGAGAAGGCTA
Ecad	ATACACTCTGGTGGTTCAGGCTGC	CCTCATTCTCAGGCACTTGACCC
Hoxb7	CTGGCGCCAAGGAGCAGAGG	CGGCCTCGCTTTCGGTCAGG
Wnt9b	GCAGGAGCGCTGGAACTGCA	CCAGTGCATGCGTGAGGGCA
mTert	AGCAAAAACCTTCCTCAGCA	AGTGAGCAGGCAGCTGGTAT

Supplementary Figure legends:

Supplementary Figure 1. A. Low resolution confocal images show the GFP⁺ tubular structures in the medulla region of neonatal kidney 4 days after injection. Scale bar: 100 μ m. **B.** High resolution confocal images demonstrate the masking and spot counting strategy performed using Imaris software for quantification of integration capacity of bulk GFP⁺ cells or PKH-26 labelled Hoxb7-derived cells. Quantification on the integration efficiency was assessed using Imaris software (Version 7.2, BitPlane AG). Separate spot counting was performed to determine the number of DAPI⁺ cells within the Aqp2⁺ structures and the number of DAPI⁺ nuclei within the GFP⁺ (or PKH26⁺) structures. Co-localisation of Aqp2 and GFP (or PKH26) was manually checked to ensure every GFP⁺ (or PKH26⁺) cell counted was also positive for Aqp2 staining. A total of 6288 Aqp2⁺ cells were counted and $12 \pm 0.8\%$ of these cells were positive for GFP. PKH26-labelled Hoxb7-derived cells accounted for $19 \pm 3.2\%$ of total 3144 Aqp2⁺ cells in the chosen regions. Scale bar 10 μ m.

Supplementary Figure 2. A. Low resolution confocal images show the localisation of GFP⁺ tubular structures in the medulla region and detection of proliferation based on positive staining for mitotic marker pHH3 (red) 4 days after injection. Arrowheads indicate occasional pHH3⁺ cells within the GFP⁺ structures. Scale bar 100 μ m. **B.** High resolution confocal images demonstrate the masking and spot counting strategy performed using Imaris software for quantification of proliferation rate of integrated GFP⁺ kidney MSCs and native collecting duct epithelial cells (Hoxb7/EGFP⁺) in postnatal day 6 kidney. A total of 4780 GFP⁺DAPI⁺ nuclei were counted using Imaris and $1.2 \pm 0.2\%$ of these nuclei were double positive for pHH3 and GFP. Analysis for normal proliferation rate of collecting duct epithelial cells using the Hoxb7/EGFP mice showed that $0.5 \pm 0.2\%$ (total 3044 nuclei counted) of Hoxb7/EGFP⁺

cells were pHH3⁺ positive in a normal postnatal day 6 kidney (comparable age). Scale bar 10µm.

Supplementary Figure 3. No overlap between macrophages and GFP after neonatal injection of kidney derived-stromal cells. Immunofluorescence staining for the macrophage marker F4/80 (red), GFP (green) and DAPI (blue) in kidney sections at day 14 post-delivery of GFP⁺ kidney-derived stromal cells. Arrowhead indicates F4/80⁺ cells close to a GFP⁺ tubular structure. Scale bar: 50µm.

Supplementary Figure 4. (A) FACS plot showing the results of sorting from total adult kidney to generate fractions for MSC culture. Lin⁻CD31⁻ cells were sorted into three fractions based on expression of CD24 and Sca-1 and collected for further analysis. MSC-like cultures could only be generated from the CD31⁻Lin⁻CD24^{lo}Sca1⁺ fraction. (B) FACS plot showing freshly isolated EGFP⁺ (Hoxb7⁺) cells from the Hoxb7/EGFP mice has low level of CD24 expression (CD24^{lo}).

Supplementary Figure 5. Graph showing the comparison of population doubling time (PDTs) between Hoxb7-derived and bulk cultured kidney MSC-like cells at passage 3, 5, 9 and 13. At passage 3 the average population doubling time is 3.1 days for Hoxb7-derived MSCs and 5.0 days for bulk cultured MSCs.

Supplementary Figure 6. Lack of F4/80 expression in the Hoxb7GFP^{lo} fraction. (A) Reanalysis of the total kidney F4/80⁺ fraction for GFP vs EpCAM reveals no overlap between the GFP⁺ population and the macrophage population. (B) Immunofluorescence for Hoxb7GFP (green), F4/80 (red) and DAPI (blue) confirms no spatial overlap between F4/80⁺

cells and either the GFP⁺ or GFP^{lo} (arrowhead) population of the PND5 kidney. Scale bar: 20µm.

Supplementary Figure 7. *In situ* hybridization of TS28 (PND6) mouse kidney (GUDMAP:14084; www.gudmap.org) show *Wnt4* expression (arrowhead) is seen in the interstitial compartment of the medulla but also in occasional collecting duct epithelial cells in the tip of the papilla at PND6. Scale bar: 100µm.

Supplementary Figure 8. (A) Low resolution confocal images show *Wnt4* (EGFP) and TdTomato (red) distribution in cortex, medulla, papilla base and tip of the adult kidney (PND49). Scale bar: 20 µm. (B) Semi-quantification data show the number of TdTomato⁺ (and EGFP⁺) cells either within Aqp2⁺ (collecting duct epithelial) or Aqp2⁻ population, confirming no EGFP⁺ (TdTomato⁺) cells are detected within the collecting duct epithelial at E19.5 but at PND49 8% of Aqp2⁺ cells are TdTomato⁺ (EGFP⁺).

Supplementary Figure 9. Evidence for convergent extension in the elongation of the neonatal collecting duct. Immunofluorescence for GFP (green) and DAPI (blue), showing Hoxb7GFP⁺ tubular structures in PND0 and PND15 mouse kidney. Dashed line outlines the edges of each tubule. Scale bar: 20µm.

Supplementary Table 1. List of primers used for qPCR.