Figure S1. The extent of glandular injury and regeneration induced by ligation and deligation of Wharton's duct and periductal tissues.

(A) Low magnification images of H&E stained sections of non-injured and glands injured by ligation of Wharton's duct for 3 days and collected at 0, 5, 10 and 20 days after deligation.
(B-C) Representative fluorescent images of control, injured and regenerative glands stained for Aqp5 or Mist1. (D) Images of control (c) and injured (L) submandibular glands at 20 days post-ligation. Graph shows mean gland weight ±s.d. for 3 glands/group. P=0.064 using t-test. E) H&E stained sections of regenerative SMGs when ligation was extended to 7 days. Images are representative of n=3 mice. Scale bars=50 μm.

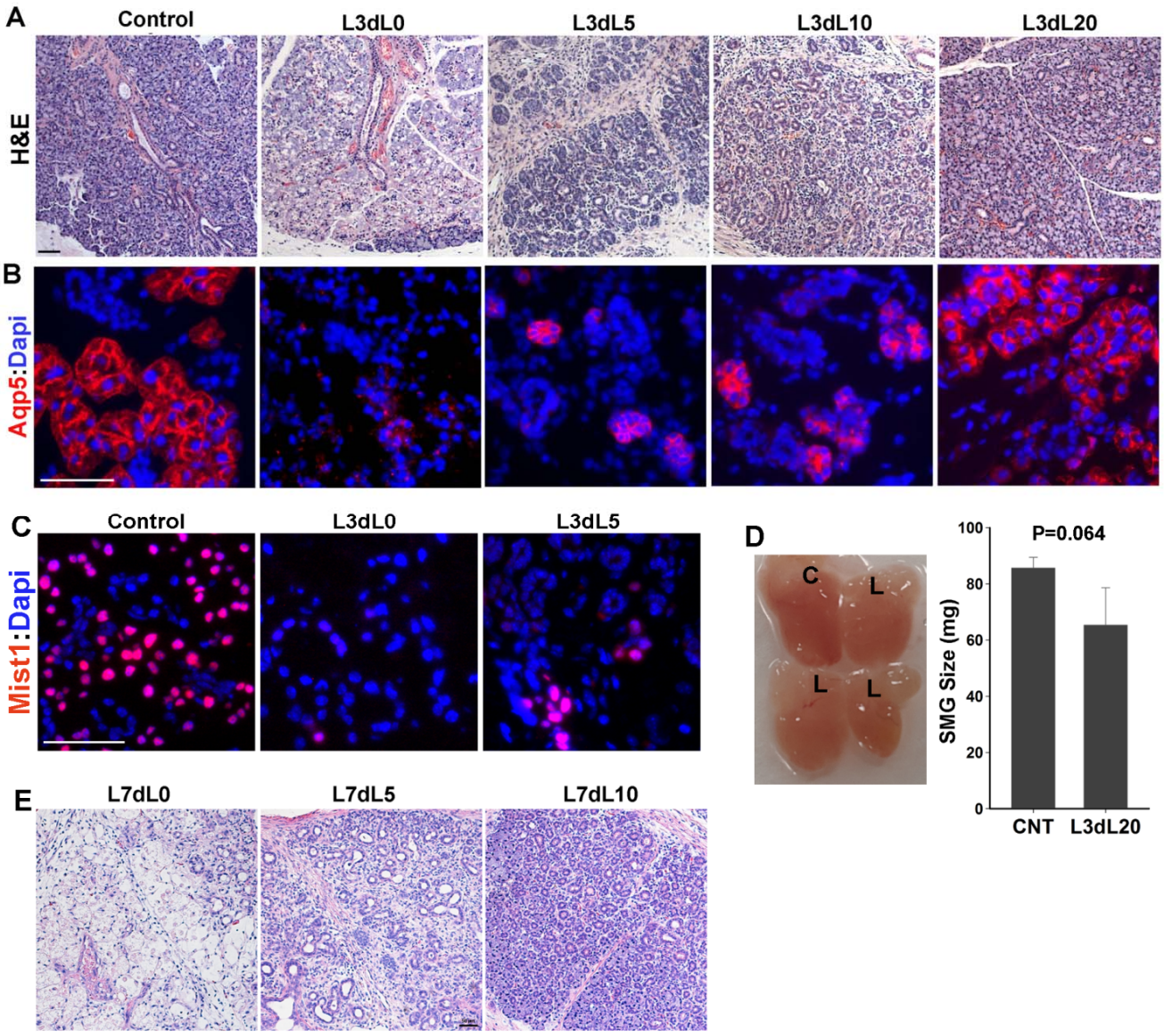

Figure S2. Distribution of nuclear H2BGFP in the control and regenerative glands collected at 10 days post-ligation. (A) Fluorescent images of H2B GFP-labeled control and regenerative glands at low magnification. (B-E) Images of control and regenerative glands (L3dL10) stained for K14 (B), SMA (C), Aqp5 (D) and cKit (E). Arrowhead in B control points to ductal stem cell cluster. In E, arrow points to MECs/basal cells surrounding cKit^{+} ID cells and arrowheads point to dim nuclear GFP in cKit^{+} cells. Images of representative of n=3 mice. Scale Bars=50 um. (F) Graph shows the percentage of GFP-labeled Aqp5^{+} and cKit^{+} cells. Data is presented as mean±sd for n=3 glands and there is no comparison between the two groups.

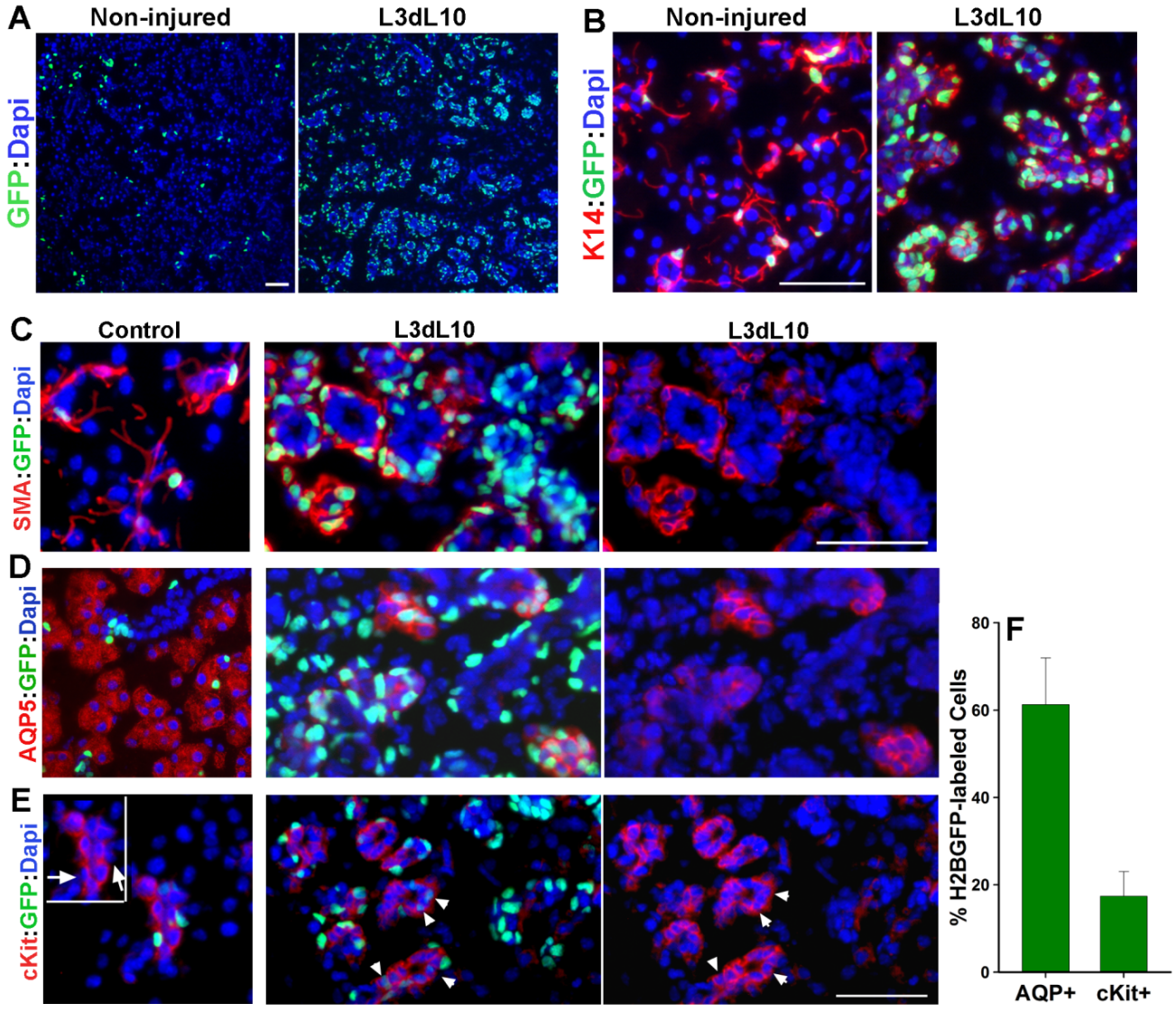

Figure S3. Mapping of K14-YFP-lineage-labeled cells in regenerated SMG ligated for 3 days and collected at 4 weeks post-deligation.
(A-B) Fluorescent image of lineage labeled clusters stained for K14 (A) or K19 (B) in red and YFP in green. Arrow in A points to ductal stem cells and inset is higher magnification of YFP-labeled stem cells. In B merged and single channels are shown. Nuclei are stained with dapi. (C) A representative image of regenerated acini (Ac) and associated intercalated ducts (ID) stained for cKit (green) and YFP (red). Arrows point to the acini-ID junction. Both merged and single channel images are shown. (D) Regenerated SMG sections were stained for SMA (red) and YFP (green). Merged and single channels images are shown. Arrows indicated unlabeled MECs and arrowhead points to YFP-labeled MEC surrounding lineage-traced acini. The bottom panel show YFP-labeled MECs surrounding non-labeled areas in the regenerated gland. Scale bars=50 um. Data is representative of n=3 glands.

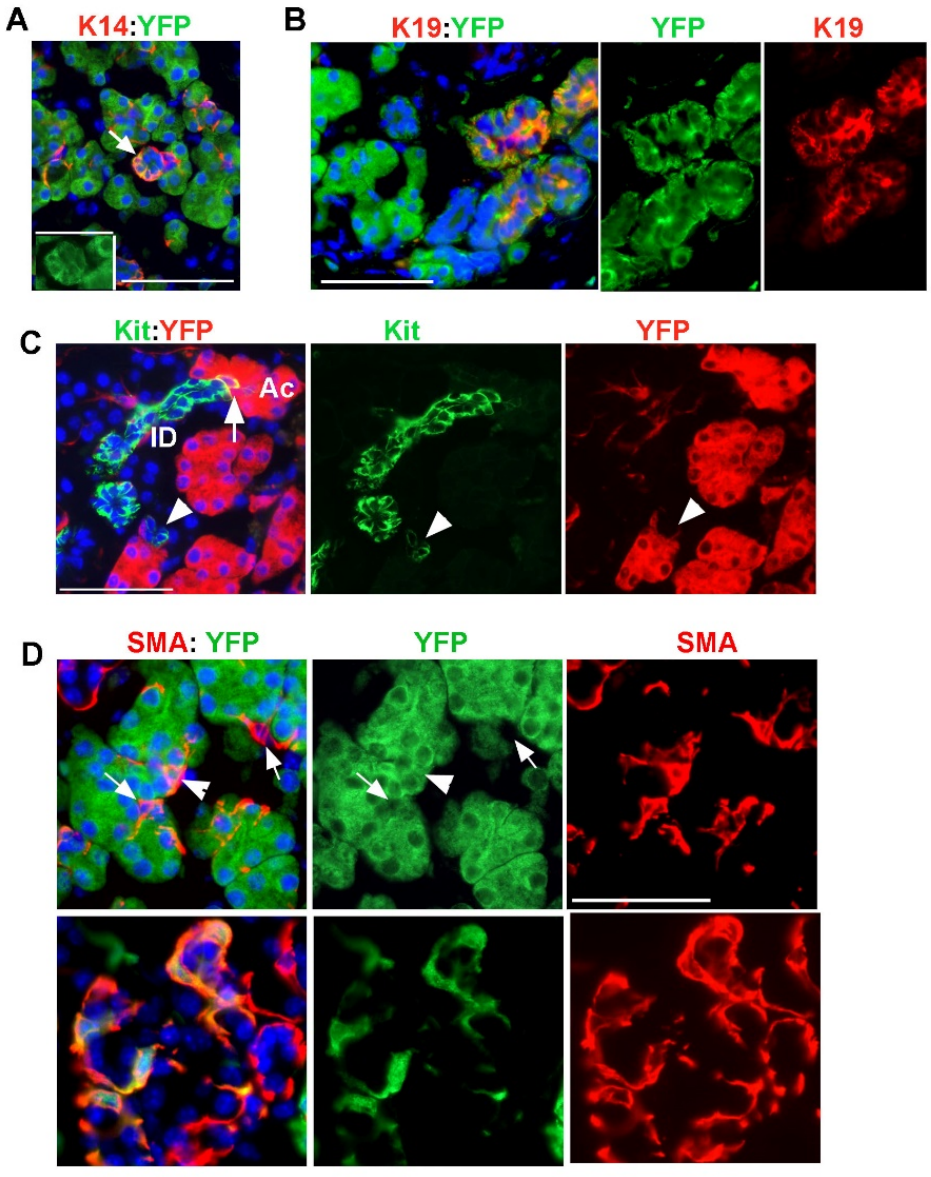

Figure S4-Lineage tracing of MECs in control and regenerated SMG

Schematics represent time line of tamoxifen administration and duct ligation and deligation. The contralateral gland was used as control. (A-F) Immunofluorescent images of control SMGs collected from *αSMA-Cre^{ERT2}:R26R-TdT* at 9 wks after Tam administration and stained with antibodies against phenotypic markers including: SMA (MECs), Aqp5 (acinar cells),cKit (ID cells) and K14 (ductal stem cells and MECs). (B-G) Images of regenerated glands at 4 wks post-deligation stained for phenotypic markers including SMA, Aqp5, cKit and K19 (granular duct cells). Red is TdT direct fluorescence. Nuclei are stained with dapi (blue). Scale Bars=50 μm. Data is representative of 3 glands.

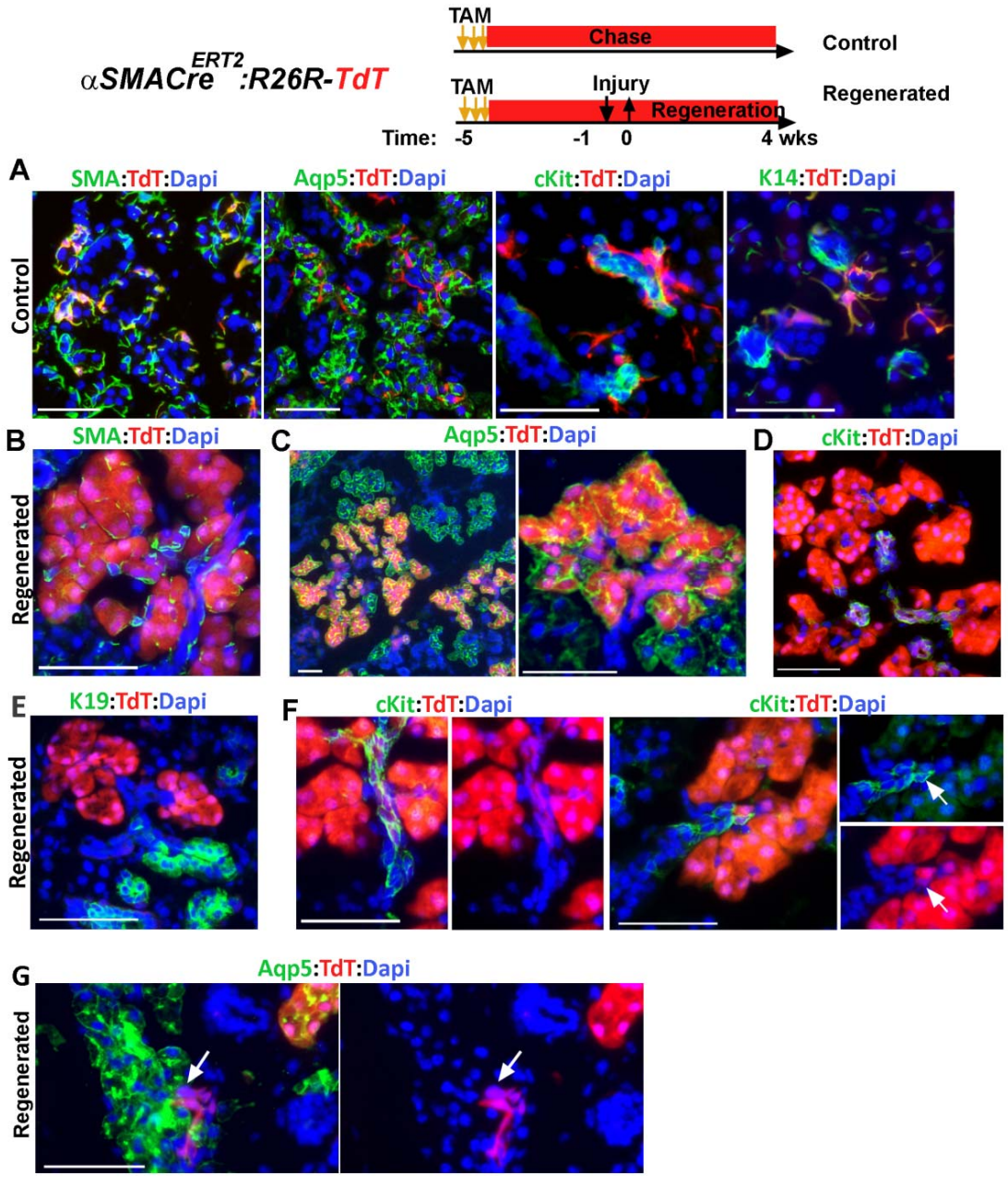

Figure S5. Comparative analysis of lineage traced cells in K14-TdT and Axin2-TdT mice.
The time line of TAM-induction of TdT and duct ligation and deligation in *Axin2Cre^{ERT2}:R26R-TdT* (Axin2-TdT) and *K14Cre^{TRE}:R26RTdT* (K14-TdT) is the same as Fig. 4A.
(A-B) Images of control SMG sections from each transgenic line stained for Aqp5 or SMA. (C-D) Images of regenerated SMGs from both transgenic mice stained for Aqp5. (D) Image of regenerated glands of Axin2-TdT mice stained for SMA. Arrows note lack of TdT expression in SMA^{+} cells. TdT is red and nuclear blue is dapi. Data is representative of 2 mice. Scale Bars=50μm.

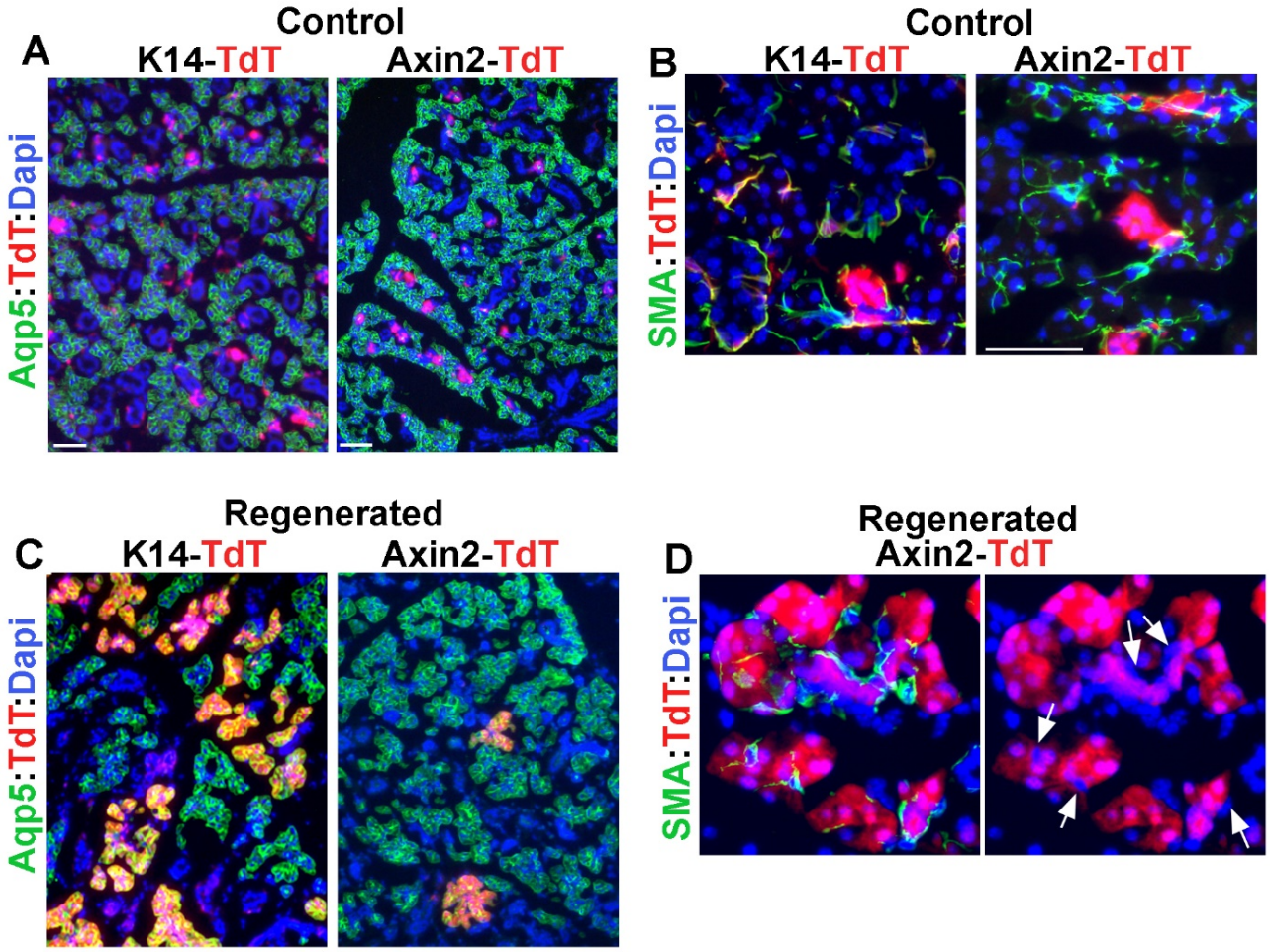

Figure S6. Lineage tracing of cKit$^+$ ID cells in the control and regenerated SMG of *cKitCreERT2:R26RTdT* mice.

Experimental design is described in Fig. 6. (A)Fluorescent images of control glands stained for cKit (ID) or Mist1 (acini). (B) Graph showing higher labeling efficiency of cKit$^+$ cells in Kit-TdT mice when compared with those for K14$^+$ and SMA$^+$ cells in K14-YFP and SMA-TdT mice, respectively. Data is presented as mean±SD for 3 mice/group, ** P<0.01, NS, not significant, one way ANOVA with post-hoc Tukey test. (C-G) Fluorescent images of regenerated SMG stained with the indicated antibody to phenotypic markers (green). Red is TdT and nuclear blue staining is dapi. In E, higher magnification of the boxed area is shown. Arrowhead in D points to ductal stem cells and arrows in F-G point to Ac/ID junction. Data represents 3 regenerated glands. Scale bars= 50 μm.

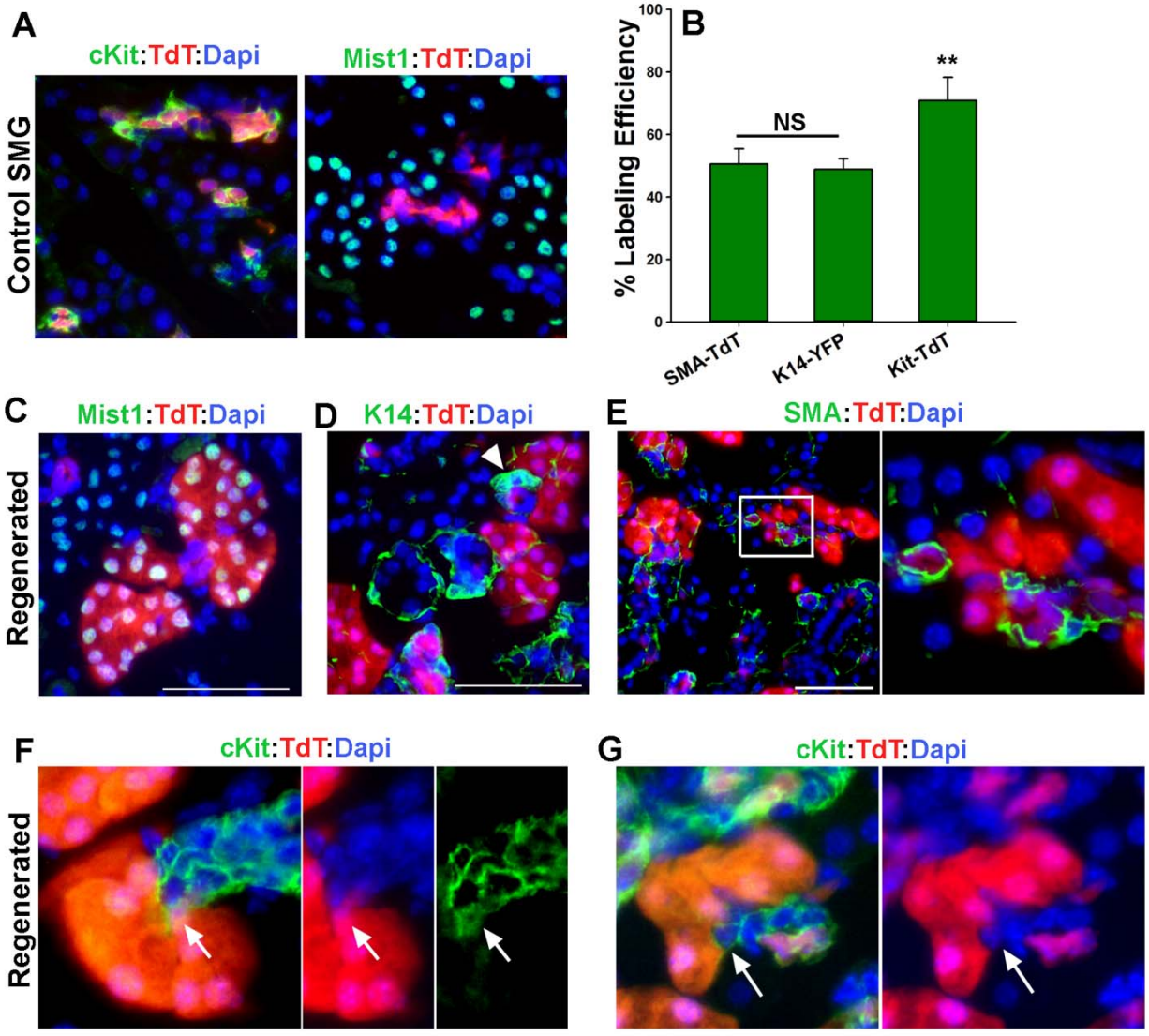

Figure S7. Comparative analysis of SMG in two model of ligation-induced injury.
(A) Images of control and injured gland either after ligation of Wharton's duct for 7 days (Duct only) or ligation of duct/periductal tissue for 3 days. Sections were stained for Aqp5. Nuclear blue is dapi. Arrow points to Aqp^{+} cells in the periphery of the gland and inset is at higher magnification. (B) Quantification of $Aqp5^{+}$ in injured glands relative to the control. Data presented as mean±s.d. (n=2 mice/group, 4 random 10X images) $P<0.001$ using one way ANOVA with post-hoc Tukey test (C) Distribution of cKit-lineage traced cells in control and regenerated glands at 4 weeks post deligation in two models of ligation-induced injury. Sections were stained for Aqp5 (green) and TdT is direct fluorescence (red). TdT^{+} cells in the larger ducts are tuft cells. Nuclear Blue is dapi. Scale Bar=100μm.

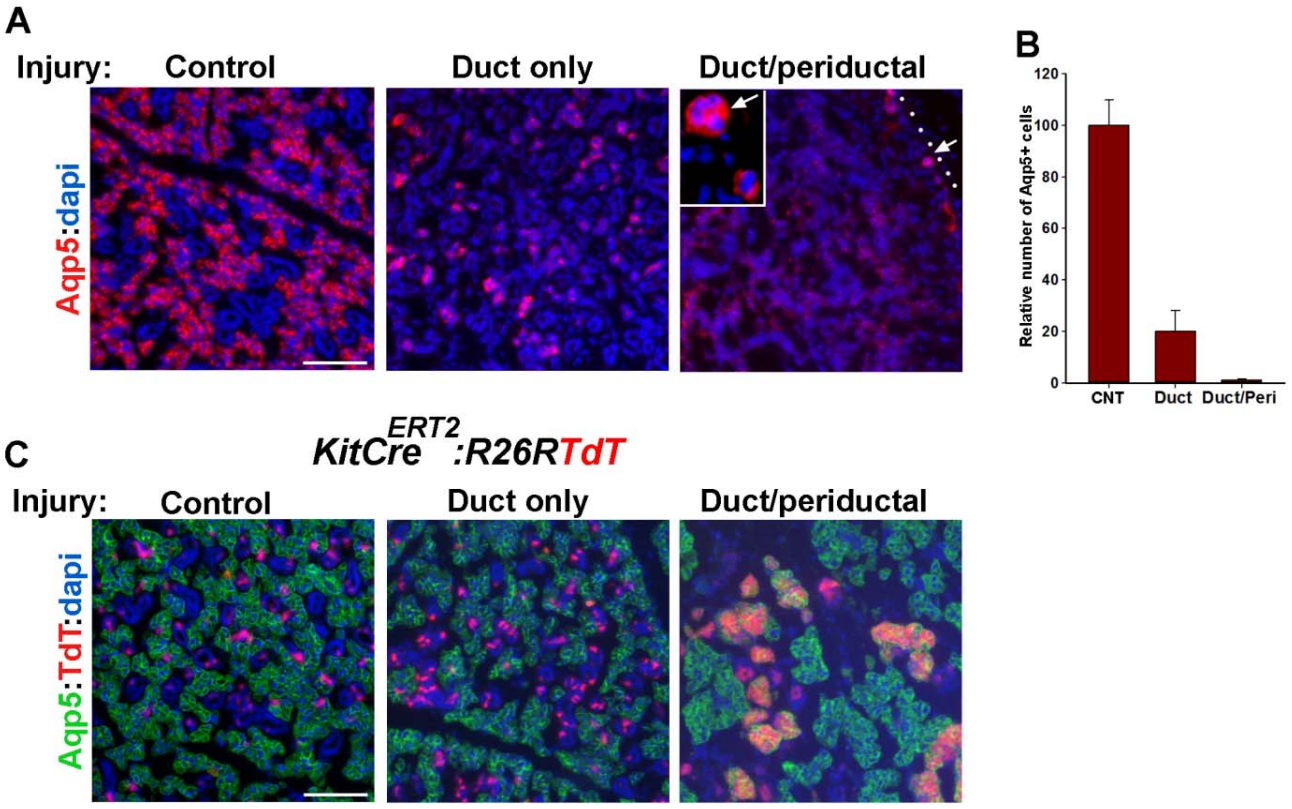

Figure S8. Sox10 expression in normal adult SMG. Images of normal 8 wk old female FVB mouse co-stained for Sox10 (nuclear green) and either Aqp5 or SMA. The dotted line shows boundaries of an ID. Blue numclear stain is dapi. ID is intercalated duct and GD is granular duct. Scale bar=50 μm.