SUPPLEMENTAL METHODS

RNA was isolated using the RNeasy isolation system (QIAGEN, Valencia, CA) and reverse transcribed using the iScript^{TM} cDNA synthesis kit (Bio-Rad, Hercules, CA). Equal amounts of cDNA reactions were amplified with FastStart Universal SYBR Green Master (Rox) (Roche) using the primer sets listed in Supplemental Table IV. Target sequences were amplified using 95^{o}C/30 sec, 59^{o}C/60 sec, 72^{o}C/30 sec conditions. Fold changes in gene expression between TM-treated and control animals were calculated using the comparative $2^{-\Delta\Delta Ct}$ method. Samples were internally normalized to 18S ribosomal RNA, $\Delta Ct = Ct_{sample} - Ct_{18S}$. Samples were then normalized to untreated controls, $\Delta\Delta Ct = \Delta Ct_{TM\text{-}treated} - \Delta Ct_{untreated}$. The inverse log of the $\Delta\Delta Ct$ was then calculated to give the relative fold change. Error bars represent the S.E.M.

SUPPLEMENTAL TABLE I
GENOTYPING AND CRE-INDUCED RECOMBINATION PRIMER SETS

Mice were genotyped using the following primer sets:

$Xbp1^{fl/+}$	5'-acttgcaccaacacttgccatttc-3', 5'-caaggtggttcactgcct-3'
$Mist1^{CreER/+}$	5'-ggttaaagcaaattgtcaagtacgg-3', 5'- atagtaagtatgtgcgtcagcg-3', 5'-gaagcattttccaggtatgctcag-3'
$R26^{LacZ/+}$	5'-gcgaagagtttgtcctcaacc-3', 5'- ggagcgggagaaatggatatg-3', 5'-aaagtcgctctgagttgttat-3'

$Xbp1^{fl/+}$ recombination was detected using the following primer sets:

$Xbp1^{WT}$	5'-ttgggactctctcgtgtg-3', 5'-caaggtggttcactgcct-3'
$Xbp1^{\Delta Ex2}$	5'-tggccacgtctacaaatgaa-3', 5'-caaggtggttcactgcct-3'

SUPPLEMENTAL TABLE II
PRIMARY ANTIBODIES FOR IMMUNOHISTOCHEMISTRY

anti-Amylase	171534, 1:100, Calbiochem, San Diego, CA
anti-β-gal	NB100-65209, 1:300, Novus Biologicals, Littleton, CO
anti-Chop	2895S, 1:500, Cell Signaling, Boston, MA
anti-E-Cadherin	ab53033, 1:1000, Abcam, Cambridge, MA
anti-Glucagon	A0565, Dako, Carpinteria, CA
anti-Ki67	M7249, 1:500, Dako, Carpinteria, CA
anti-K19	Troma3, 1:100, Dev. Studies Hybridoma Bank, Iowa City, IA
anti-Mist1	C175, 1:500, Pin et al, 2000
anti-pH3	06-570, 1:100, Upstate (Millipore), Billerica, MA
anti-LC3B	3868S, 1:200, Cell Signaling, Boston, MA
anti-Sox9	AB5535, 1:500, Millipore, Billerica, MA
anti-Hes1	Gift of Tetsuo Sudo, 1:1000
anti-F4/80	MF48000, 1:50, Invitrogen, Camarillo, CA
anti-CD3	A0452, 1:200, Dako, Carpinteria, CA

SUPPLEMENTAL TABLE III
PRIMARY ANTIBODIES FOR IMMUNOBLOTS

anti-nATF6α	sc-22799, 1:1000, Santa Cruz Biotechnology, Santa Cruz, CA
anti-p-eIF2α	3597S, 1:1000, Cell Signaling, Boston, MA
anti-Amylase	sc-12821, 1:1000, Santa Cruz Biotechnology, Santa Cruz, CA
anti-CPA	1810-0006, 1:1000, AbD Serotec, Raleigh, NC
anti-Erk1/2	9102S, 1:1000, Cell Signaling, Boston, MA
anti-p-Erk1/2	9101S, 1:1000, Cell Signaling, Boston, MA
anti-p38	9212S, 1:1000, Cell Signaling, Boston, MA
anti-S6	sc-74459, 1:1000, Santa Cruz Biotechnology, Santa Cruz, CA

SUPPLEMENTAL TABLE IV
RT-QPCR PRIMER SETS

$Xbp1^{\Delta Ex2}$	5'-agaaagcgctgcggagaac-3', 5'-cctccacctctggaacctc-3'
Bip	5'-gtgtcctctctggtgatcagg-3', 5'-tgtcttttgttaggggtcgtt-3'
Chop	5'-cctgaggagagagtgttccag-3', 5'-cagatcctcataccaggcttc-3'
Xbp1u	5'-tcagactatgtgcacctctgc-3', 5'-agagaaagggaggctggtaag-3'
Xbp1s	5'-tgagtccgcagcaggt-3', 5'-agagaaagggaggctggtaag-3'
Sec61a	5'-ctatttccagggctccgagt-3', 5'-aggtgtgtactggcctcggt-3'
Pdi1	5'-caagatcaagccccacctgat-3', 5'-agttcgccccaaccagtactt-3'
Amylase	5'-cagagacatggtgacaaggtg-3', 5'-atcgttaaagtcccaagcaga-3'
Elastase	5'-actatgtccagctgggtgttc-3', 5'-cagtaagaggagctggagcag-3'
Nestin	5'-gagagtcgcttagaggtgcag-3', 5'-gatctgagcgatctgactctgt-3'
Hes1	5'-agagaaggcagacattctgga-3', 5'-gtcacctcgttcatgcactc-3'
Gli1	5'-tttcttgaggttgggatgaag-3', 5'-ggtggagtcattggattgaac-3'
Mist1	5'-tggtggctaaagctacgtgt-3', 5'-catagctccaggctggtttt-3'
Sox9	5'-cttctgtgggagcgacaactt-3', 5'-agggagggaaaacagagaacg-3'
Reg1	5'-atggctaggaacgcctacttc-3', 5'-cccaagttaaacggtcttcagt-3'
18S	5'-tgtctcaaagattaagccatgc-3', 5'-gcgaccaaaggaaccataac-3'

SUPPLEMENTAL TABLE V
MARKER EXPRESSION PROFILE

COMPARTMENT	MARKERS EXPRESSED
Acinar Cells	Mist1, Amylase, Carboxypeptidase (CPA)
Islets	Insulin, Glucagon
Ductal/centroacinar	Cytokeratin 19 (K19), Sox9
Centroacinar exclusive	Hes1, Nestin
All epithelial cells	E-cadherin

CONTEXT	MARKERS EXPRESSED
General ER stress	BIP, CHOP
Xbp1 Pathway	Xbp1s, Sec61a, PDI
PERK Pathway	p-eIF2α
ATF6a Pathway	nATF6α
Apoptosis	TUNEL
Actively proliferating	Ki67, phospho-histone 3 (pH3)
Cre-recombination (ROSA)	β-galactosidase
Autophagy	Autophagy marker light chain 3 - isoform B (LC3B)
Regeneration	Reg1

SUPPLEMENTAL FIGURE LEGENDS

FIGURE S1: *The Xbp1$^{fl/fl}$ locus has no effect on acinar cell morphology.* **(A,B)** H&E staining of pancreata sections from control wild type and Xbp1$^{\Delta Ex2/+}$; Mist1$^{CreER/+}$ mice treated with corn oil. All genotypes exhibit normal appearing pancreata. Scale = 40 μm. **(C,D)** Relative transcript levels of the common ER stress indicators BiP and Chop from 2 wk post-TM treated mice reveals that pancreata heterozygous for Xbp1 do not undergo an ER stress response. Only homozygous null Xbp1$^{\Delta Ex2}$ cells show ER stress responses. *p<0.05

FIGURE S2: *Islet and duct populations are normal following acinar-specific Xbp1 ablation*. **(A)** Insulin/glucagon costaining of Xbp1$^{\Delta Ex2}$ pancreata 4 wk post-TM reveal normal islet morphology. **(B)** K19 staining of Xbp1$^{\Delta Ex2}$ pancreata also indicates normal ductal development in 4 wk post-TM pancreata. Scale = 20 μm.

FIGURE S3: *Xbp1$^{fl/+}$;Mist1$^{CreER/+}$ and Xbp1$^{fl/fl}$;Mist1$^{CreER/+}$ zymogen-bearing acini have normal zymogen accumulation, localization and development of rER.* **(A,B)** Xbp1$^{fl/+}$;Mist1$^{CreER/+}$ pancreata show no alterations in zymogen granule accumulation, abortive zymogens, or ER disorganization. **(C,D)** Zymogen-containing, acinar cell populations in 4 wk post-TM Xbp1$^{fl/fl}$,Mist1$^{CreER/+}$ pancreata are similar in ultrastructure to heterozygous and wild-type controls. **(E,F)** Non-zymogenic exocrine cells in 4 wk post-TM Xbp1$^{fl/fl}$; Mist1$^{CreER/+}$ pancreata show the previously described cell abnormalities including few and mislocalized zymogens, self-destructing organelles, and disrupted ER organization. **(G,H)** Eight wk post-TM Xbp1$^{fl/fl}$; Mist1$^{CreER/+}$ pancreata

exhibit an almost full recovery from Xbp1 deletion with an increase in ZG numbers and a well developed rER. Scale = 1 μm (A,C,E,G); 0.5 μm (B,D,F,H).

FIGURE S4: *Anti-β-galactosidase staining indicates zymogenic cells fail to ablate Xbp1.* Zymogenic cells (outlines - identified under phase contrast by extensive ZG accumulation) in $Xbp1^{fl/fl}$;$Mist1^{CreER/+}$;$R26R^{LacZ}$ pancreata 4 wk post-TM do not express β-gal (green), indicating no Cre-dependent recombination. Scale = 20 μm.

FIGURE S5: *$Xbp1^{\Delta Ex2}$ cells undergo autophagy*. **(A)** LC3B staining (green) in 4 wk post-TM pancreata reveals the presence of extensive autophagic cells within the non-zymogenic cell compartment. In contrast, zymogenic cells (red outlines) and islets (yellow outline) exhibit no signs of autophagy. **(B)** Electron micrograph of a non-zymogenic acinar cell showing a zymogen granule within an autolysosome (red box). Scale = 20 μm (A); 1 μm (B). **(C)** Immunoblots for additional stress pathway components (pERK1/2 and p38) show that they are elevated in $Xbp1^{\Delta Ex2}$ pancreata over the indicated post-TM time course. As predicted, maximum MAPK stress is observed at 4 wk post-TM.

FIGURE S6: *$Xbp1^{\Delta Ex2}$ pancreata undergo extensive cell proliferation of $Xbp1^{fl/fl}$ acinar cells.* **(A)** Quantification of Ki67 positive $Xbp1^{\Delta Ex2}$ acinar cells over the indicated post-TM time course. **(B)** H&E staining reveals mitotic figures (arrow) in pancreata 6 wk post-TM. Scale = 10 μm. **(C)** Relative transcript levels of Reg1 over the indicated post-TM time points. *$p<0.05$

FIGURE S7: *Xbp1$^{\Delta Ex2}$ pancreata reveal rare β-gal+ acinar cells 8 wk post-TM.* Anti-β-gal staining of 8 wk post-TM Xbp1$^{\Delta Ex2}$ pancreata shows a rare intact acinus (white outline) that is β-gal positive, presumably reflecting cells that recombined the R26^{LacZ} locus but failed to delete both copies of the Xbp1$^{fl/fl}$ allele. Scale = 10 μm.

FIGURE S8: *Recovered Xbp1$^{\Delta Ex2}$ pancreata contain areas of tubular duct-like structures surrounded by stromal cells.* **(A)** High-magnification image of tubular duct complexes (arrows) in 8 wk Xbp1$^{\Delta Ex2}$ pancreata. **(B,C)** Surrounding the tubular complexes (TC) are CD3+ T cells and F4/80+ macrophages (arrows). Ac - acinar cells; BV - blood vessel. Scale = 20 μm.

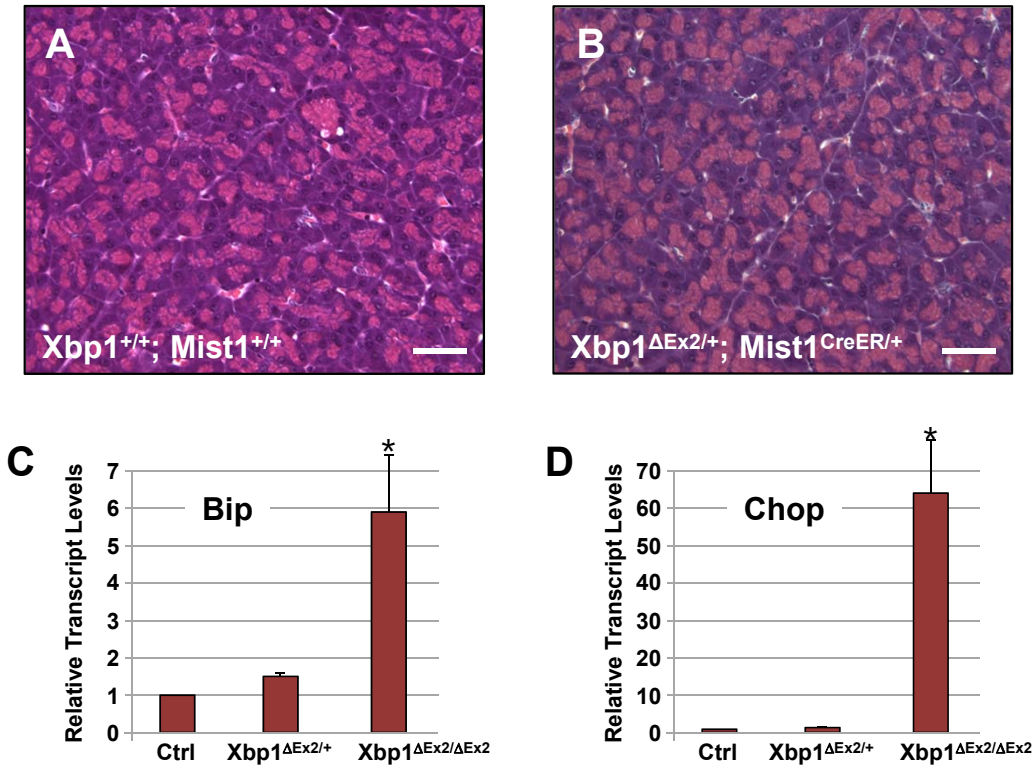

A
Xbp1+/+; Mist1+/+
B
Xbp1ΔEx2/+; Mist1CreER/+
C
Bip
Relative Transcript Levels
0
1
2
3
4
5
6
7
*
Ctrl
Xbp1ΔEx2/+
Xbp1ΔEx2/ΔEx2
D
Chop
Relative Transcript Levels
0
10
20
30
40
50
60
70
*
Ctrl
Xbp1ΔEx2/+
Xbp1ΔEx2/ΔEx2

Figure S1

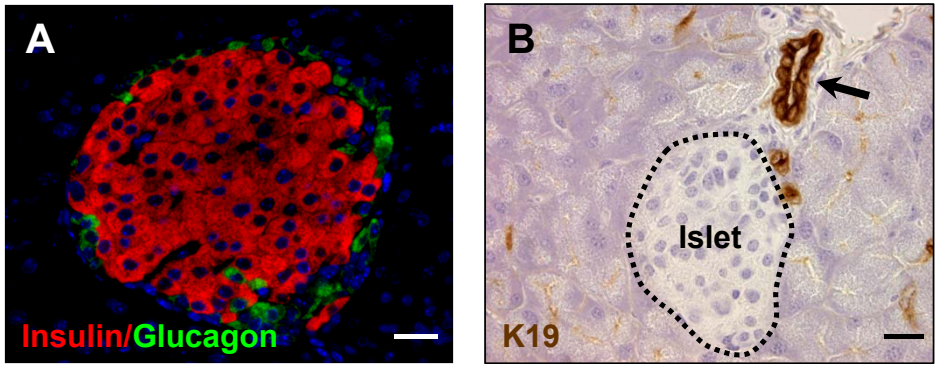


Figure S2

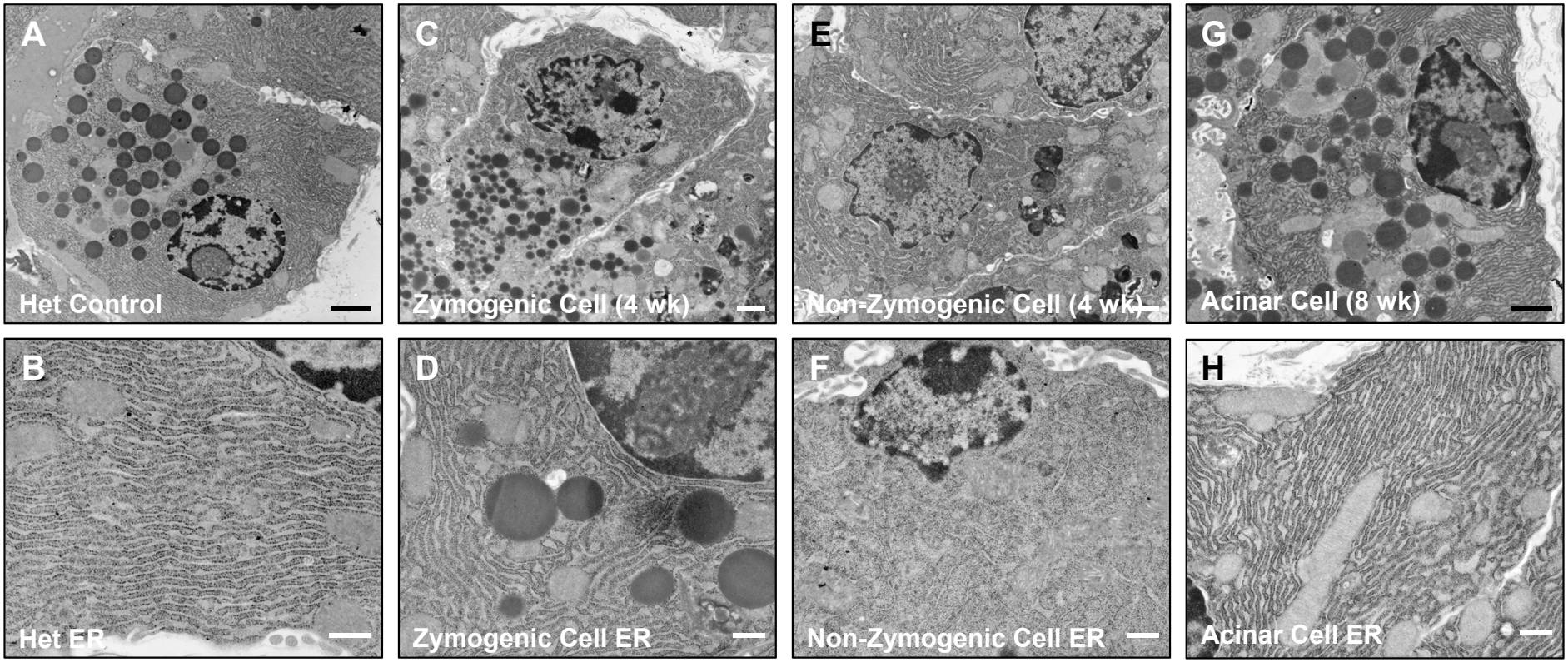

A
Het Control
C
Zymogenic Cell (4 wk)
E
Non-Zymogenic Cell (4 wk)
G
Acinar Cell (8 wk)
B
Het ER
D
Zymogenic Cell ER
F
Non-Zymogenic Cell ER
H
Acinar Cell ER

Figure S3

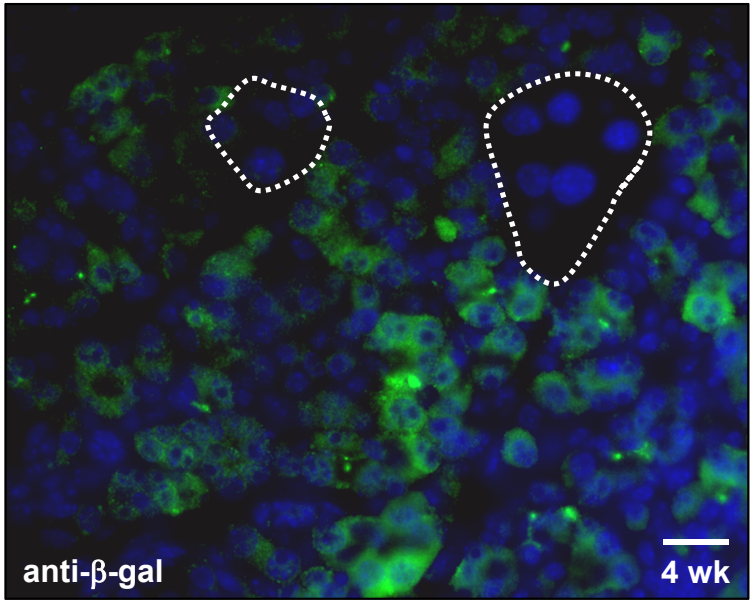
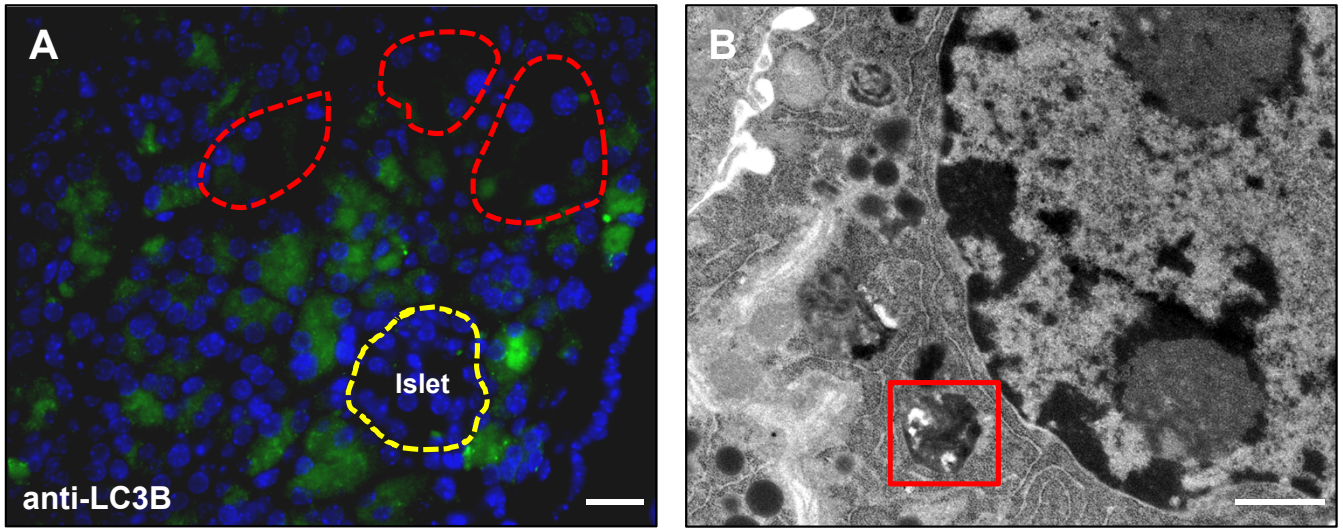


Figure S4



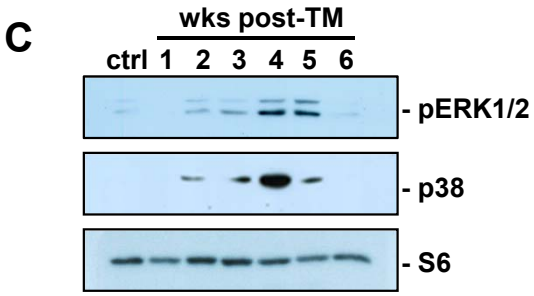
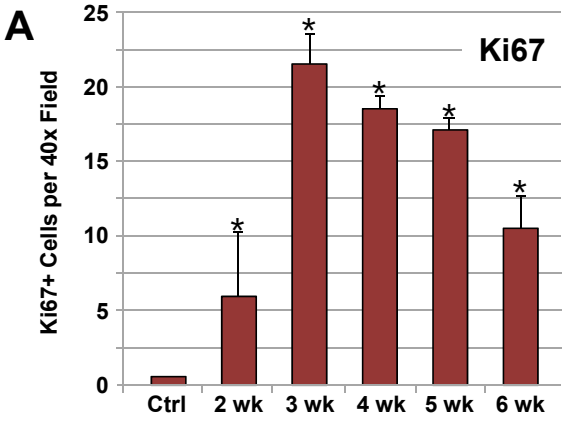
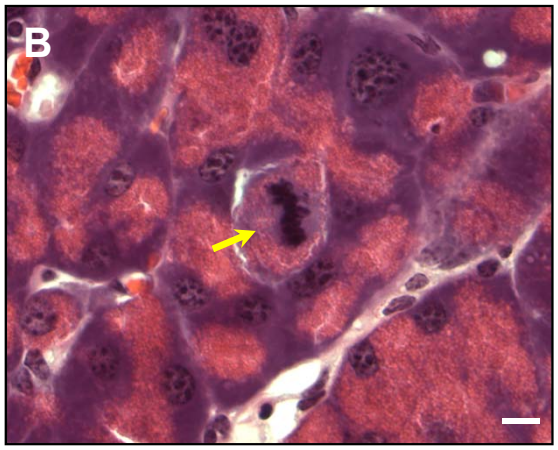


Figure S5


A
Ki67
Ki67+ Cells per 40x Field
25
20
15
10
5
0
Ctrl
2 wk
3 wk
4 wk
5 wk
6 wk


B

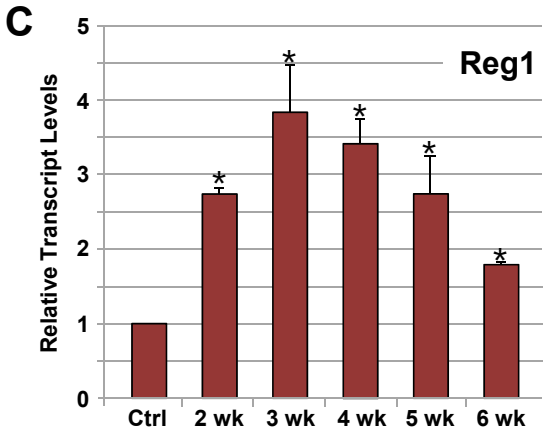

C
Reg1
Relative Transcript Levels
5
4
3
2
1
0
Ctrl
2 wk
3 wk
4 wk
5 wk
6 wk

Figure S6

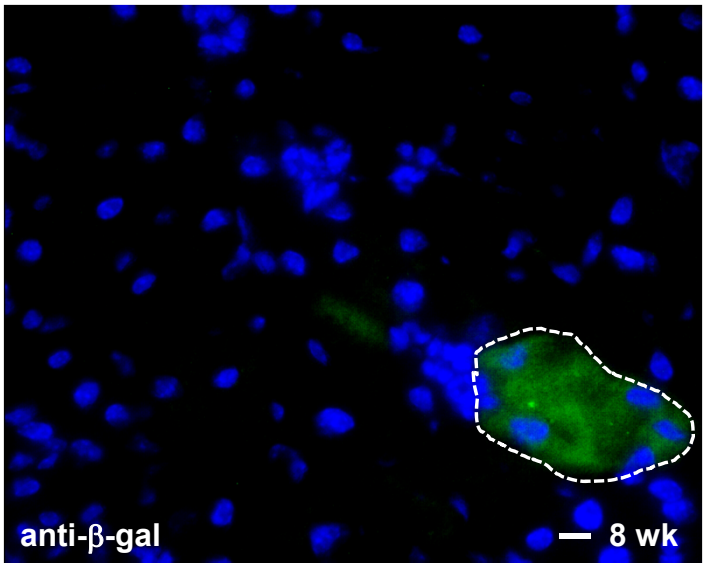


Figure S7

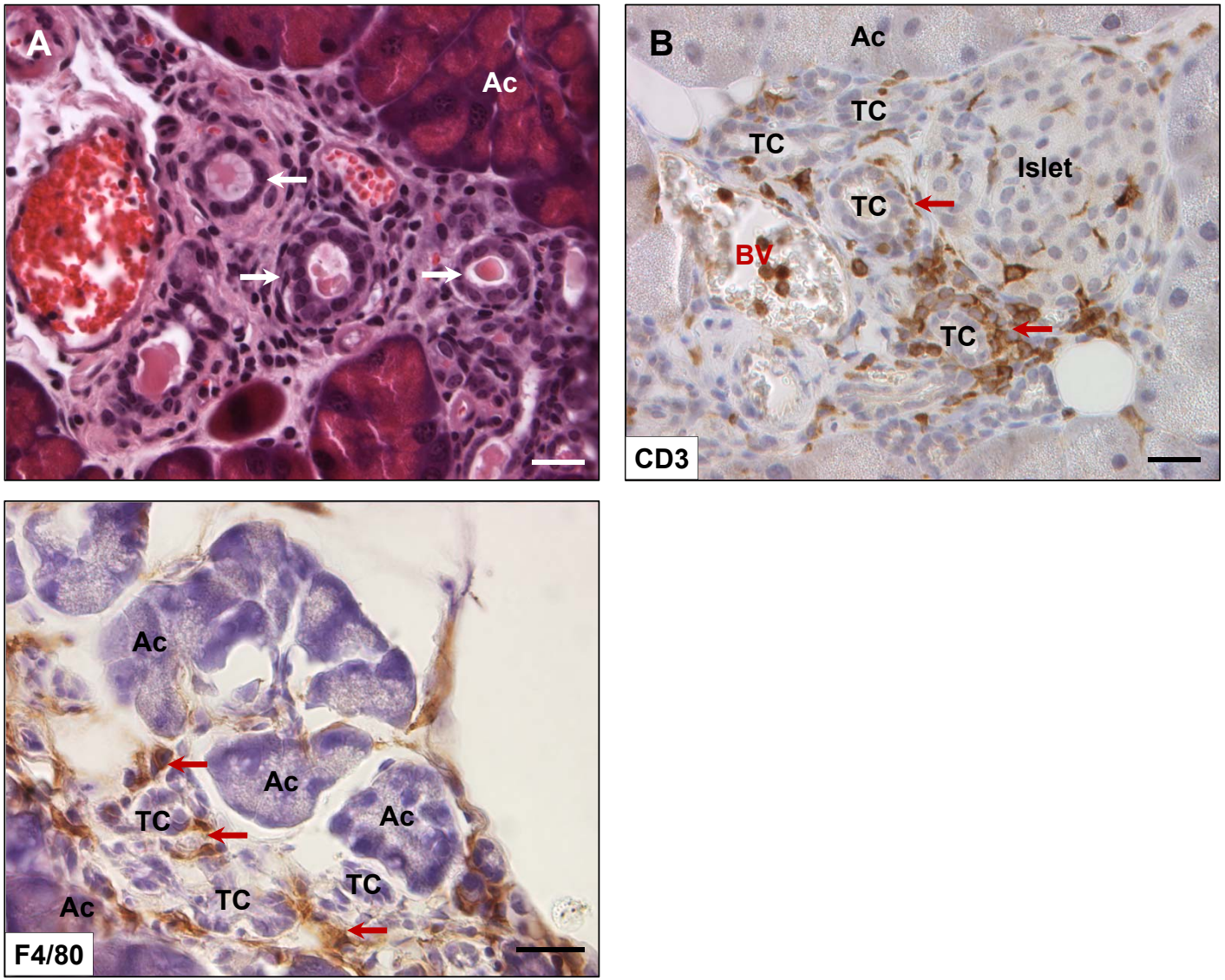


Figure S8