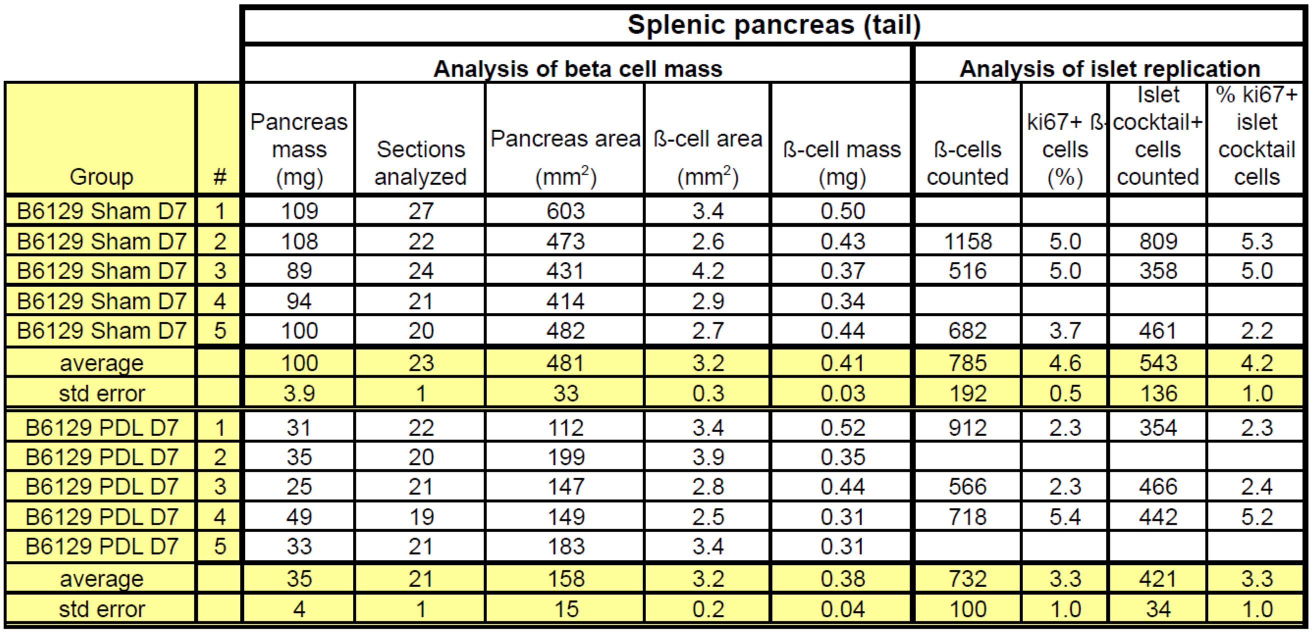
SUPPLEMENTARY DATA

Supplementary Table 1. Individual data from sham-operated or PDL 7 day mixed genetic background cohort at 7 days. (top) Age, body weight, and glycemia of sham-operated and PDL cohort. (middle) Morphometry of pancreas head. (bottom) Morphometry and proliferation analysis of pancreas tail.

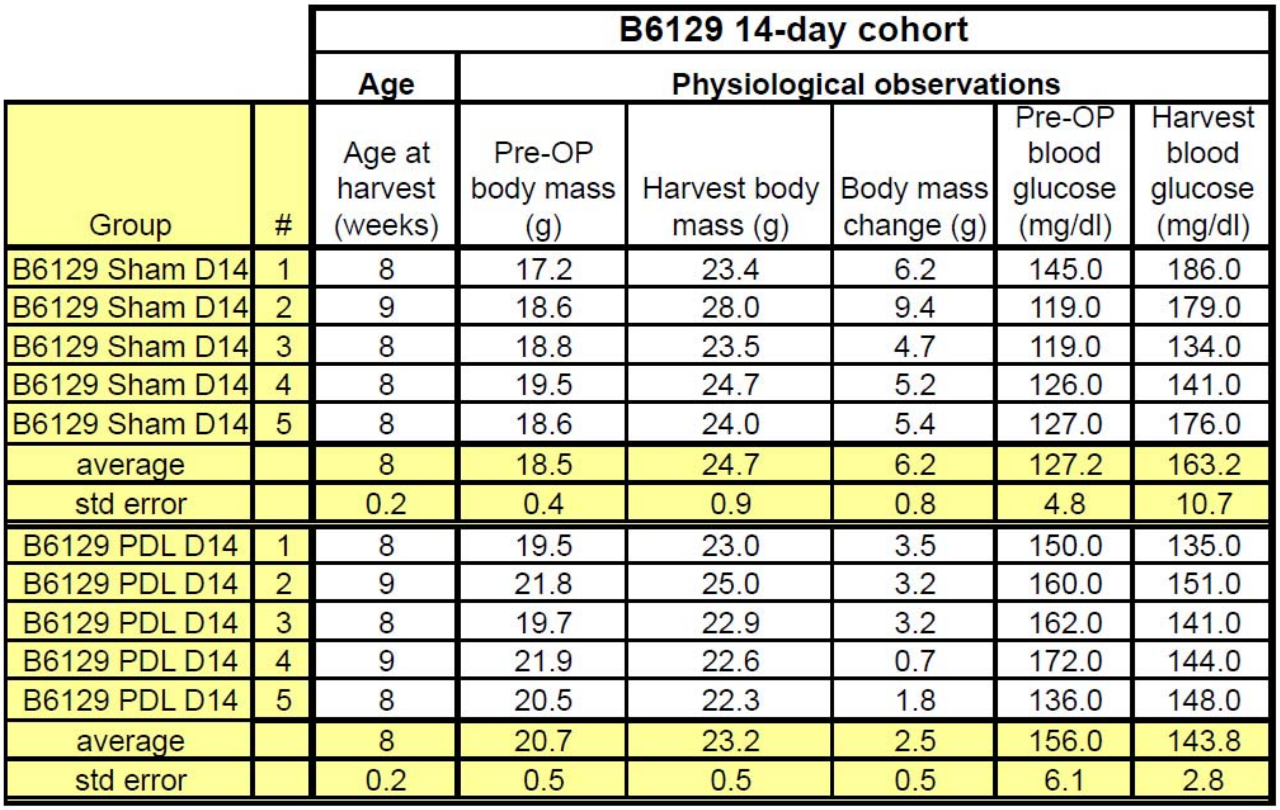
		B6129 7-day cohort					
		Age	**Physiological observations**				
Group	#	Age at harvest (weeks)	Pre-OP body mass (g)	Harvest body mass (g)	Body mass change (g)	Pre-OP blood glucose (mg/dl)	Harvest blood glucose (mg/dl)
B6129 Sham D7	1	7	20.0	21.4	1.4	154.0	129.0
B6129 Sham D7	2	7	19.5	21.8	2.3	150.0	130.0
B6129 Sham D7	3	7	14.7	20.8	6.1	148.0	153.0
B6129 Sham D7	4	7	16.4	21.6	5.2	171.0	112.0
B6129 Sham D7	5	7	17.4	21.8	4.4	158.0	146.0
average		7	17.6	21.5	3.9	156.2	134.0
std error		0.0	1.0	0.2	0.9	4.1	7.2
B6129 PDL D7	1	8	21.6	22.1	0.5	154.0	141.0
B6129 PDL D7	2	7	17.4	20.7	3.3	155.0	160.0
B6129 PDL D7	3	7	18.5	19.0	0.5	171.0	133.0
B6129 PDL D7	4	7	18.2	19.4	1.2	147.0	154.0
B6129 PDL D7	5	7	21.0	21.8	0.8	156.0	142.0
average		7	19.3	20.6	1.3	156.6	146.0
std error		0.2	0.8	0.6	0.5	3.9	4.8

		Duodenal and gastric pancreas (head)				
		Analysis of beta cell mass				
Group	#	Pancreas mass (mg)	Sections analyzed	Pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)
B6129 Sham D7	1	83	27	552	3.3	0.50
B6129 Sham D7	2	95	26	647	2.9	0.43
B6129 Sham D7	3	80	24	355	1.6	0.37
B6129 Sham D7	4	75	22	307	1.4	0.34
B6129 Sham D7	5	101	17	385	1.7	0.44
average		87	23	449	2.2	0.41
std error		4.8	2	64	0.4	0.03
B6129 PDL D7	1	102	30	333	1.7	0.52
B6129 PDL D7	2	72	25	384	1.8	0.35
B6129 PDL D7	3	97	29	808	3.7	0.44
B6129 PDL D7	4	68	27	354	1.6	0.31
B6129 PDL D7	5	81	25	366	1.4	0.31
average		84	27	449	2.0	0.38
std error		6.7	1	90	0.4	0.04



		Splenic pancreas (tail)								
		Analysis of beta cell mass					Analysis of islet replication			
Group	#	Pancreas mass (mg)	Sections analyzed	Pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)	ß-cells counted	ki67+ ß-cells (%)	Islet cocktail+ cells counted	% ki67+ islet cocktail cells
B6129 Sham D7	1	109	27	603	3.4	0.50				
B6129 Sham D7	2	108	22	473	2.6	0.43	1158	5.0	809	5.3
B6129 Sham D7	3	89	24	431	4.2	0.37	516	5.0	358	5.0
B6129 Sham D7	4	94	21	414	2.9	0.34				
B6129 Sham D7	5	100	20	482	2.7	0.44	682	3.7	461	2.2
average		100	23	481	3.2	0.41	785	4.6	543	4.2
std error		3.9	1	33	0.3	0.03	192	0.5	136	1.0
B6129 PDL D7	1	31	22	112	3.4	0.52	912	2.3	354	2.3
B6129 PDL D7	2	35	20	199	3.9	0.35				
B6129 PDL D7	3	25	21	147	2.8	0.44	566	2.3	466	2.4
B6129 PDL D7	4	49	19	149	2.5	0.31	718	5.4	442	5.2
B6129 PDL D7	5	33	21	183	3.4	0.31				
average		35	21	158	3.2	0.38	732	3.3	421	3.3
std error		4	1	15	0.2	0.04	100	1.0	34	1.0

Supplementary Table 2. Individual data from sham-operated or PDL 14 day mixed genetic background cohort at 7 days. (top) Age, body weight, and glycemia of sham-operated and PDL cohort. (middle) Morphometry of pancreas head. (bottom) Morphometry and proliferation analysis of pancreas tail.



		B6129 14-day cohort					
		Age	Physiological observations				
Group	#	Age at harvest (weeks)	Pre-OP body mass (g)	Harvest body mass (g)	Body mass change (g)	Pre-OP blood glucose (mg/dl)	Harvest blood glucose (mg/dl)
B6129 Sham D14	1	8	17.2	23.4	6.2	145.0	186.0
B6129 Sham D14	2	9	18.6	28.0	9.4	119.0	179.0
B6129 Sham D14	3	8	18.8	23.5	4.7	119.0	134.0
B6129 Sham D14	4	8	19.5	24.7	5.2	126.0	141.0
B6129 Sham D14	5	8	18.6	24.0	5.4	127.0	176.0
average		8	18.5	24.7	6.2	127.2	163.2
std error		0.2	0.4	0.9	0.8	4.8	10.7
B6129 PDL D14	1	8	19.5	23.0	3.5	150.0	135.0
B6129 PDL D14	2	9	21.8	25.0	3.2	160.0	151.0
B6129 PDL D14	3	8	19.7	22.9	3.2	162.0	141.0
B6129 PDL D14	4	9	21.9	22.6	0.7	172.0	144.0
B6129 PDL D14	5	8	20.5	22.3	1.8	136.0	148.0
average		8	20.7	23.2	2.5	156.0	143.8
std error		0.2	0.5	0.5	0.5	6.1	2.8

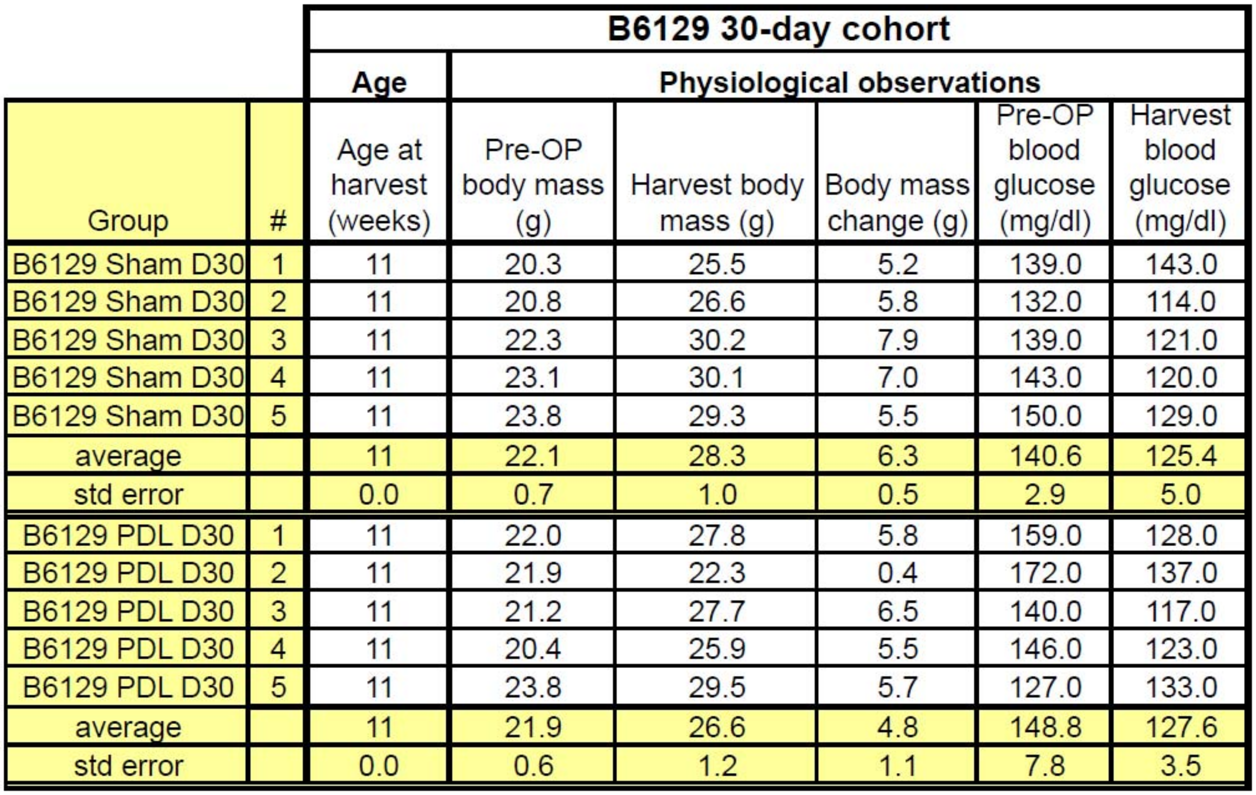
SUPPLEMENTARY DATA

		Duodenal and gastric pancreas (head)				
		Analysis of beta cell mass				
Group	#	Pancreas mass (mg)	Sections analyzed	Pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)
B6129 Sham D14	1	118	28	533	1.7	0.39
B6129 Sham D14	2	113	25	545	2.5	0.52
B6129 Sham D14	3	112	0	0	0.0	
B6129 Sham D14	4	117	21	415	2.1	0.63
B6129 Sham D14	5	134	30	518	1.8	0.48
average		119	21	503	2.0	0.50
std error		4.0	5	30	0.2	0.05
B6129 PDL D14	1	106	0	0	0.0	
B6129 PDL D14	2	131	28	865	2.9	0.45
B6129 PDL D14	3	117	29	503	1.3	0.31
B6129 PDL D14	4	115	26	655	2.6	0.45
B6129 PDL D14	5	109	35	500	1.7	0.35
average		116	24	631	2.1	0.39
std error		4.3	6	86	0.4	0.03

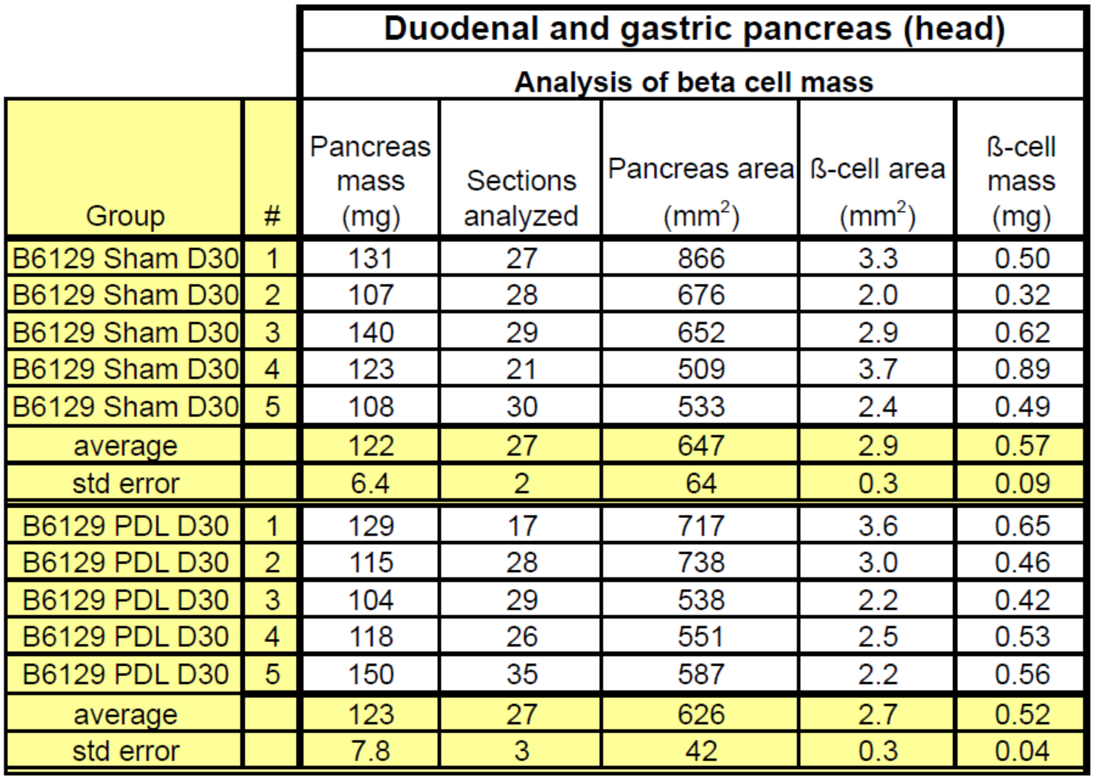
		Splenic pancreas (tail)								
		Analysis of beta cell mass					Analysis of islet replication			
Group	#	Pancreas mass (mg)	Sections analyzed	Total pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)	ß-cells counted	ki67+ ß-cells (%)	Islet cocktail+ cells counted	% ki67+ islet cocktail cells
B6129 Sham D14	1	126	36	598	4.3	0.90	778	2.18	460.295	1.30
B6129 Sham D14	2	162	28	480	5.2	0.64	913	3.3	616	1.9
B6129 Sham D14	3	129	30	595	3.4	0.73				
B6129 Sham D14	4	128	0							
B6129 Sham D14	5	132	34	465	1.9	0.55	625	2.4	289	2.4
average		135	26	534	3.7	0.70	772	2.6	455	1.9
std error		6.7	7	36	0.7	0.07	83	0.3	95	0.3
B6129 PDL D14	1	30	21	114	2.7	0.72	1268	2.4	592	3.5
B6129 PDL D14	2	29	18	171	4.0	0.68	1063	2.2	549	2.4
B6129 PDL D14	3	33	15	186	3.7	0.66				
B6129 PDL D14	4	28	15	124	3.4	0.76	684	1.6	531	1.3
average		30	17	149	3.5	0.70	1005	2.0	558	2.4
std error		1	1	18	0.3	0.02	171	0.2	18	0.6

SUPPLEMENTARY DATA

Supplementary Table 3. Individual data from sham-operated or PDL 30 day mixed genetic background cohort at 7 days. (top) Age, body weight, and glycemia of sham-operated and PDL cohort. (middle) Morphometry of pancreas head. (bottom) Morphometry and proliferation analysis of pancreas tail.



		B6129 30-day cohort						
		Age	Physiological observations					
Group	#	Age at harvest (weeks)	Pre-OP body mass (g)	Harvest body mass (g)	Body mass change (g)	Pre-OP blood glucose (mg/dl)	Harvest blood glucose (mg/dl)
B6129 Sham D30	1	11	20.3	25.5	5.2	139.0	143.0
B6129 Sham D30	2	11	20.8	26.6	5.8	132.0	114.0
B6129 Sham D30	3	11	22.3	30.2	7.9	139.0	121.0
B6129 Sham D30	4	11	23.1	30.1	7.0	143.0	120.0
B6129 Sham D30	5	11	23.8	29.3	5.5	150.0	129.0
average		11	22.1	28.3	6.3	140.6	125.4
std error		0.0	0.7	1.0	0.5	2.9	5.0
B6129 PDL D30	1	11	22.0	27.8	5.8	159.0	128.0
B6129 PDL D30	2	11	21.9	22.3	0.4	172.0	137.0
B6129 PDL D30	3	11	21.2	27.7	6.5	140.0	117.0
B6129 PDL D30	4	11	20.4	25.9	5.5	146.0	123.0
B6129 PDL D30	5	11	23.8	29.5	5.7	127.0	133.0
average		11	21.9	26.6	4.8	148.8	127.6
std error		0.0	0.6	1.2	1.1	7.8	3.5



		Duodenal and gastric pancreas (head)				
		Analysis of beta cell mass				
Group	#	Pancreas mass (mg)	Sections analyzed	Pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)
B6129 Sham D30	1	131	27	866	3.3	0.50
B6129 Sham D30	2	107	28	676	2.0	0.32
B6129 Sham D30	3	140	29	652	2.9	0.62
B6129 Sham D30	4	123	21	509	3.7	0.89
B6129 Sham D30	5	108	30	533	2.4	0.49
average		122	27	647	2.9	0.57
std error		6.4	2	64	0.3	0.09
B6129 PDL D30	1	129	17	717	3.6	0.65
B6129 PDL D30	2	115	28	738	3.0	0.46
B6129 PDL D30	3	104	29	538	2.2	0.42
B6129 PDL D30	4	118	26	551	2.5	0.53
B6129 PDL D30	5	150	35	587	2.2	0.56
average		123	27	626	2.7	0.52
std error		7.8	3	42	0.3	0.04

SUPPLEMENTARY DATA

		Splenic pancreas (tail)								
		Analysis of beta cell mass					Analysis of islet replication			
Group	#	Pancreas mass (mg)	Sections analyzed	Total pancreas area (mm^2)	ß-cell area (mm^2)	ß-cell mass (mg)	ß-cells counted	ki67+ ß-cells (%)	Islet cocktail+ cells counted	% ki67+ islet cocktail cells
B6129 Sham D30	1	135	32	638	3.5	0.74				
B6129 Sham D30	2	134	14	465	2.3	0.66				
B6129 Sham D30	3	138	28	681	4.1	0.83	191	2.1	177	0.6
B6129 Sham D30	4	130	24			0.79	654	2.8	323	1.9
B6129 Sham D30	5	138	24	785	6.0	1.06	1571	1.1	837	0.7
average		135	24	645	4.0	0.81	805	2.0	446	1.0
std error		1.5	3	52	0.6	0.07	405	0.5	200	0.4
B6129 PDL D30	1	41	13	189	3.2	0.70				
B6129 PDL D30	2	33	28	224	4.0	0.59				
B6129 PDL D30	3	60	15	228	3.9	1.04	858	2.4	471	2.1
B6129 PDL D30	4	48	17	241	3.6	0.72	1498	1.7	901	1.6
B6129 PDL D30	5	42	15			1.16	942	2.8	571.75	1.7
average		84	18	226	4.3	0.84	1099	2.3	648	1.8
std error		6.7	3	10	0.6	0.11	201	0.3	130	0.2

Supplementary Table 4. Individual data from sham-operated or PDL 7 day Balb/c cohort at 7 days. (top) Age, body weight, and glycemia of sham-operated and PDL cohort. (middle) Morphometry and proliferation analysis of pancreas head. (bottom) Morphometry and proliferation analysis of pancreas tail.

		Balb/c 7 day Cohort				
		Age	Physiological observations			
Group	#	Age at harvest (weeks)	Pre-OP body mass (g)	Harvest body mass (g)	Harvest Blood glucose (mg/dl)	Body mass change (g)
Balb/c Sham	1	9.0	27.9	29	136.0	0.6
Balb/c Sham	2	9.0	26.5	26	130.0	-0.5
Balb/c Sham	3	9.0	26.9	26	115.0	-0.8
Balb/c Sham	4	9.0	25.5	27	126.0	1.2
Balb/c Sham	5	9.0	29.6	30	121.0	0.4
average			27.3	27	125.6	0.2
std error			0.7	0.8	3.6	0.4
Balb/c PDL	1	9.0	24.1	25	127.0	1.0
Balb/c PDL	2	9.0	26.6	27	130.0	0.5
Balb/c PDL	3	9.0	25.8	26	123.0	-0.3
Balb/c PDL	4	9.0	26.0	29	124.0	2.5
Balb/c PDL	5	9.0	28.0	29	131.0	1.3
average			26.1	27	127.0	1.0
std error			0.6	0.8	1.6	0.5

SUPPLEMENTARY DATA

		Duodenal and gastric pancreas (head)						
		Analysis of beta cell mass				Analysis of islet replication		
Group	#	Pancreas mass (mg)	Sections analyzed	ß-cell area (%)	ß-cell mass (mg)	ß-cells counted	% BrdU^{+} ß-cells	% BrdU^{+} intra-islet cells
Balb/c Sham	1	159.0	19.0	0.4	0.6	2919	0.1	0.3
Balb/c Sham	2	147.0	14.0	0.4	0.6	2744	0.1	0.3
Balb/c Sham	3	133.0	13.0	0.5	0.7	2293	0.3	0.4
Balb/c Sham	4	135.0	15.0	0.5	0.6	4581	0.2	0.7
Balb/c Sham	5	153.0	15.0	0.5	0.8	3563	0.1	0.3
average		145.4	15.2	0.5	0.7	3220	0.1	0.4
std error		5.0	1.0	0.0	0.0	396.6	0.0	0.1
Balb/c PDL	1	140.0	20.0	0.3	0.5	5396	0.1	0.3
Balb/c PDL	2	129.0	15.0	0.4	0.5	2520	0.2	0.3
Balb/c PDL	3	153.0	18.0	0.3	0.5	692	0.2	0.6
Balb/c PDL	4	132.0	17.0	0.5	0.6	5832	0.2	0.3
Balb/c PDL	5	144.0	18.0	0.4	0.5	2953	0.1	0.5
average		139.6	17.6	0.4	0.5	3479	0.2	0.4
std error		4.3	0.8	0.0	0.0	953.3	0.0	0.1

		Splenic pancreas (tail)						
		Analysis of beta cell mass				Analysis of islet replication		
Group	#	pancreas mass (mg)	sections analyzed	ß-cell area (%)	ß-cell mass (mg)	ß-cells counted	% BrdU^{+} ß-cells	% BrdU^{+} intra-islet cells
Balb/c Sham	1	163.0	18.0	0.5	0.8	7544	0.1	0.5
Balb/c Sham	2	151.0	14.0	0.6	1.0	6261	0.2	0.8
Balb/c Sham	3	136.0	15.0	0.7	0.9	4280	0.4	0.5
Balb/c Sham	4	139.0	16.0	0.6	0.8	4514	0.3	0.6
Balb/c Sham	5	168.0	16.0	0.6	1.0	7189	0.4	0.8
average		151.4	15.8	0.6	0.9	5958	0.3	0.6
std error		6.3	0.7	0.0	0.0	671.7	0.1	0.1
Balb/c PDL	1	38.0	9.0	2.8	1.0	5644	0.5	1.5
Balb/c PDL	2	32.0	9.0	2.0	0.7	9297	0.3	0.9
Balb/c PDL	3	37.0	14.0	1.8	0.7	4061	0.3	1.0
Balb/c PDL	4	36.0	17.0	3.0	1.1	2716	0.7	1.7
Balb/c PDL	5	30.0	11.0	3.6	1.1	3130	0.4	1.5
average		34.6	12.0	2.6	0.9	4970	0.4	1.3
std error		1.5	1.5	0.3	0.1	1193.1	0.1	0.2

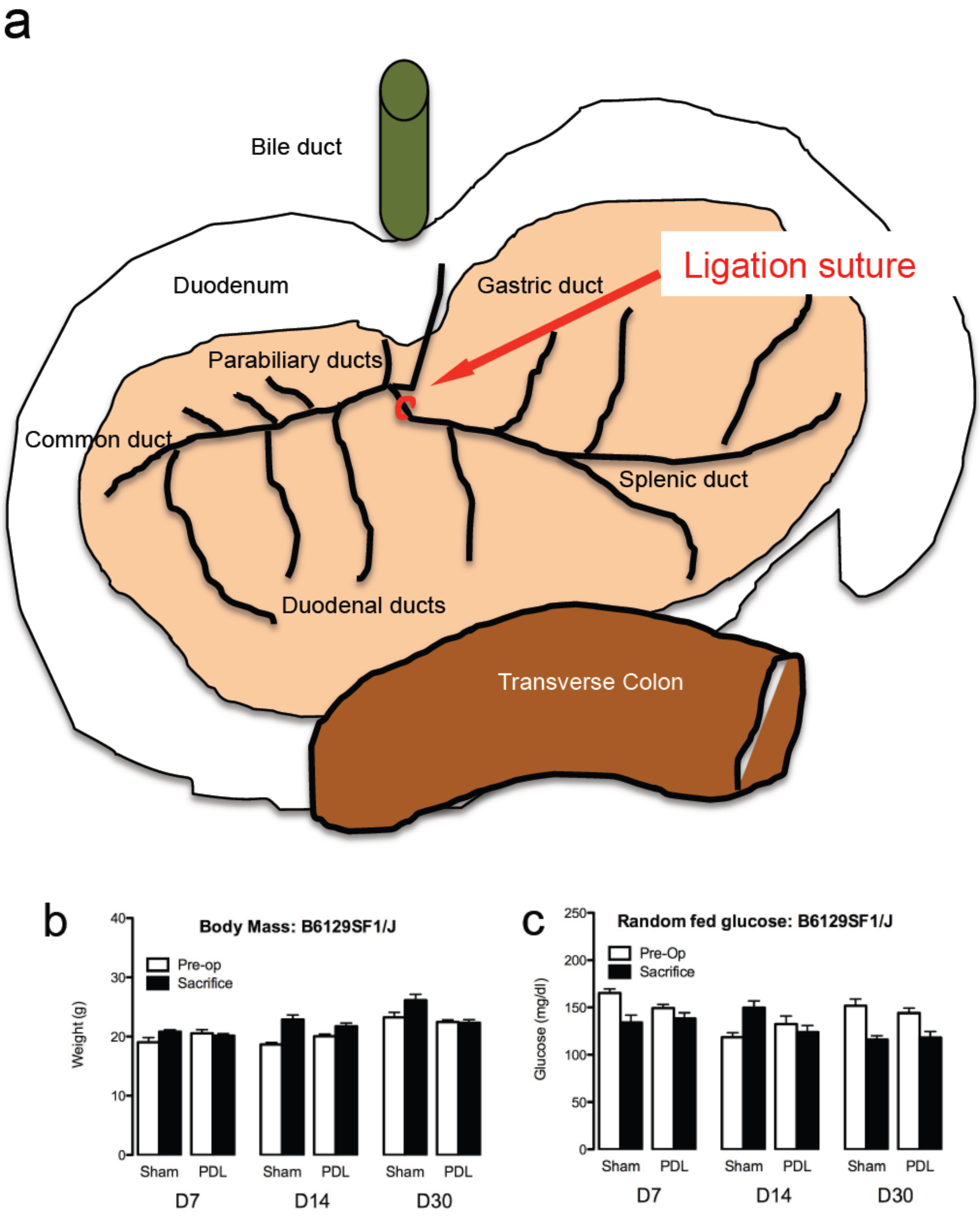
Supplementary Table 5. Primer sequences (top) and antisera (bottom).

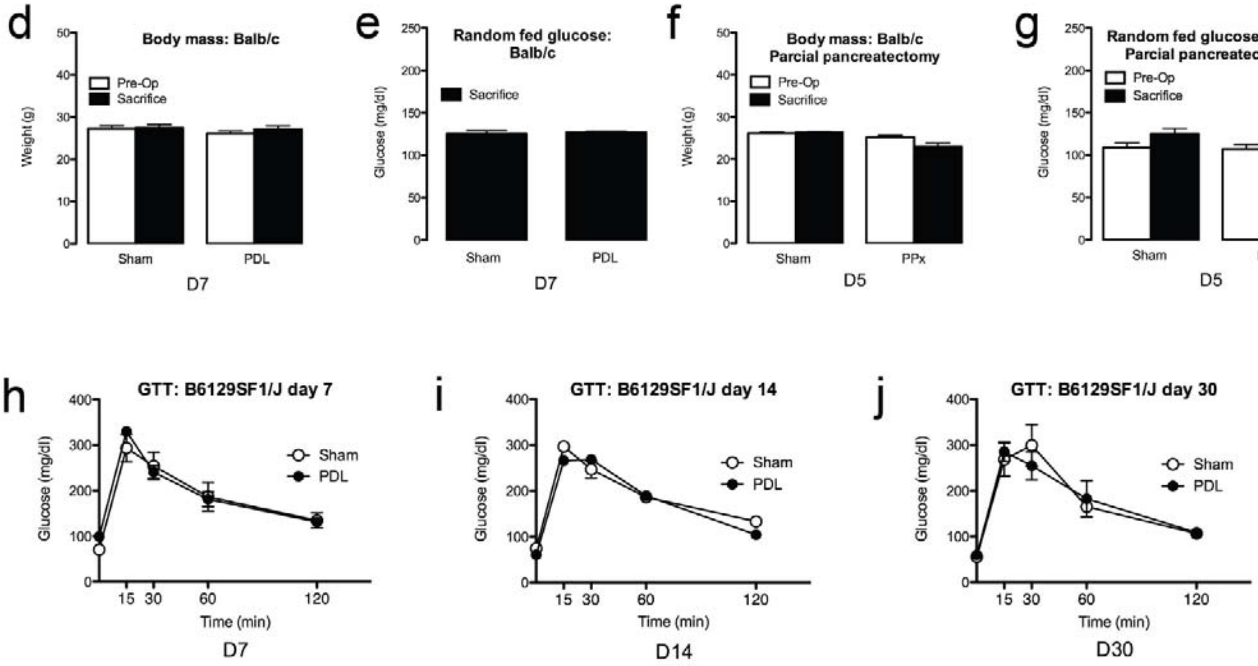
Gene	Forward	Reverse	Probe
Amylase	AAATCTGCACAAGGTCTGGA	CCACAGGTACTGCTTGTTCC	CGGACACCAACATTGTTGCACCT
Insulin	CTTCAGACCTTGGCGTTGGA	ATGCTGGTGCAGCACTGATC	CCCGGCAGAAGCGTGGCATT
Glucagon	GAATGAAGACAAACGCCACTCA	CGGCGGGAGTCGAGGTA	AGGGCACATTCACCAGCGACTACAGC
Reg3 beta	CCTGCTCCGTCATGTCCT	CGTGCGGAGGGTATATTCTT	TGCTGCTCTCCTGCCTGATGC
Reg3 gamma	ACCATCACCATCATGTCCTG	GGCATCTTTCTTGGCAACTT	TGCTGCTCTCCTGCCTGATGC
Glut2	TCTTCACGGATGCCAATTACC	AGAACTGCTGGGCCATGTG	AGCCCATCCTCGTGGCGCTG
PCSK2	TGCATAAAGACGGAGAGGAA	GGTGTAGGCTGCGTCTTCTT	TGGCCTTTGCAAGCCCATTG
IAAPP	CACACAACGCCTGGCAAAC	TGTATTCGATCCCACGTTGGT	TCGTTCCAGCAACAACCTTGGTCCA
ptp1b	CTTCCGTGGACATCAAGAAAGTACT	GGCGCCCTCGATGACA	CTCATCCAGACTGCCGACCAGCTG
Ngn3	GTCGGGAGAACTAGGATGGC	GGAGCAGTCCCTAGGTATG	CCGGAGCCTCGGACCACGAA
Nkx6.1	GCCCGGAGTGATGCAGAGT	TCTTCCCATCTTTGTCCAACAA	TGGAGGGACGCGCGCCTT
Pdx1	AGGAAAACAAGAGGACCCGTACT	CGGGAGATGTATTTGTTAAATAAGAATTC	CCTACACCCGGGCGCAGCTG
Fas	GTGGATCTGGGCTGTCCT	TCCTTGATATAATCCTTCTGAGCA	TTCACGAACCCGCCTCCTCA
FasLig	CTGGTTGGAATGGGATTAGG	TTAAGGCTTTGGTTGGTGAA	CCACCTGCAGAAGGAACTGGCA
IL1 beta	TCAGGCAGGCAGTATCACTC	CCATGAGTCACAGAGGATGG	CCCGTGGACCTTCCAGGATGA
CD45	CAAACTTCTGGCCTTTGGAT	CACTGGGTGTAGGTGTTTGC	TGCCCTTCTGGACACAGAAGTCTTTG
F4/80	TTTGGCTGCTCCTCTTCTG	TCATTCACACCACCAAGTGTT	CCACACTTCCCACCCTGGGAC
Cyclophilin	CAGACGCCACTGTCGCTTT	TGTCTTTGGAACTTTGTCTGCAA	CCCTTGGGCCGCGTCTCCTT

Antigen	Supplier	Source	Stock
Amylase	Abcam	Rabbit	ab21156
Insulin	Zymed	Guinea Pig	4367
ki67	BD	Mouse	550609
Cytokeratin	DAKO	Rabbit	Z062201-2
CD45	BD Pharmingen	Rat	550539
Ngn3	IOWA DSHB	Mouse	F25A1B3
CldU/BrdU	Accurate Chemical	Rat	OBT0030
IdU	BD Pharmingen	Mouse	NC9516321
Gluc/PP/Som	Zymed	Rabbit	
YFP	Abcam	Rabbit	ab6673

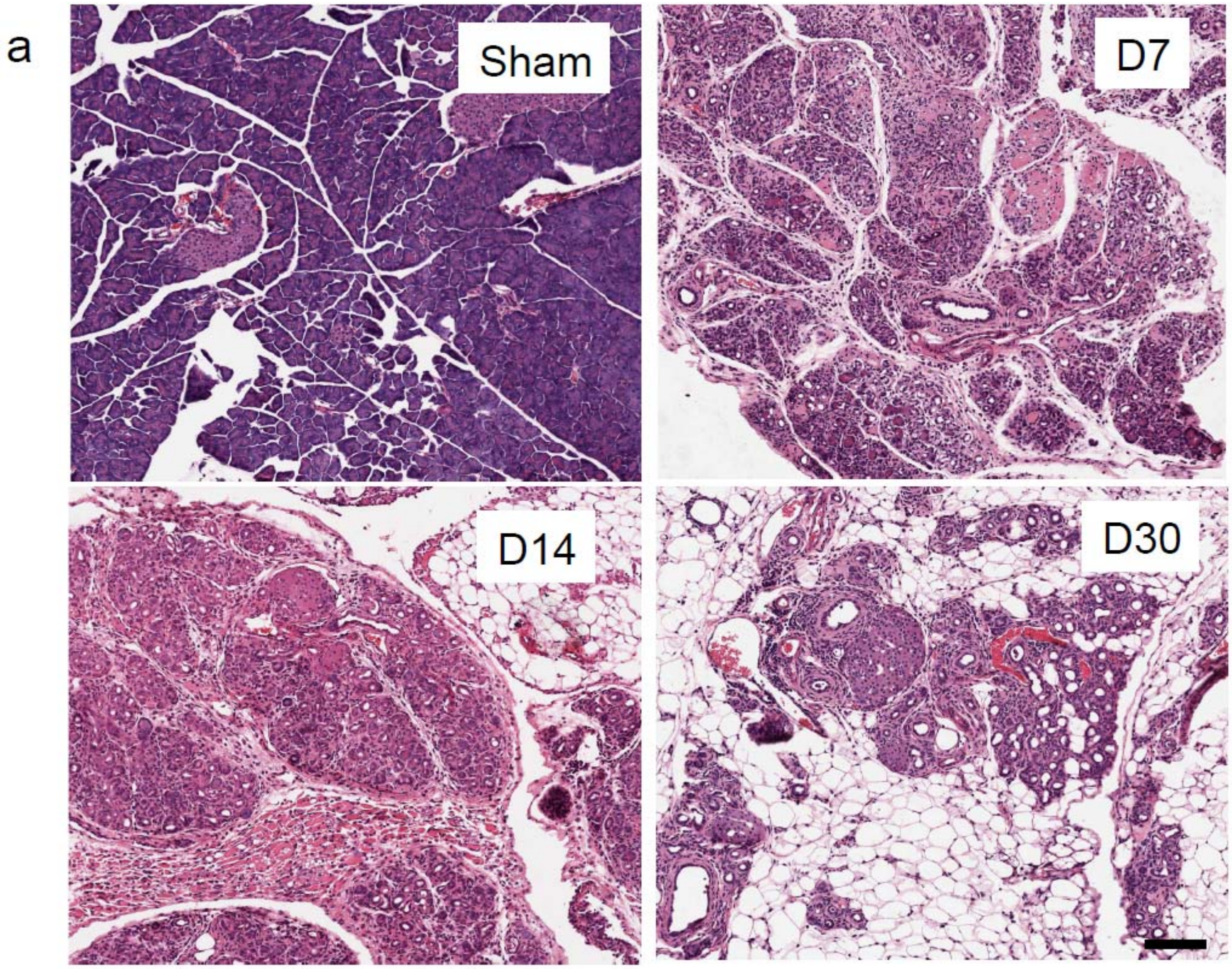
SUPPLEMENTARY DATA

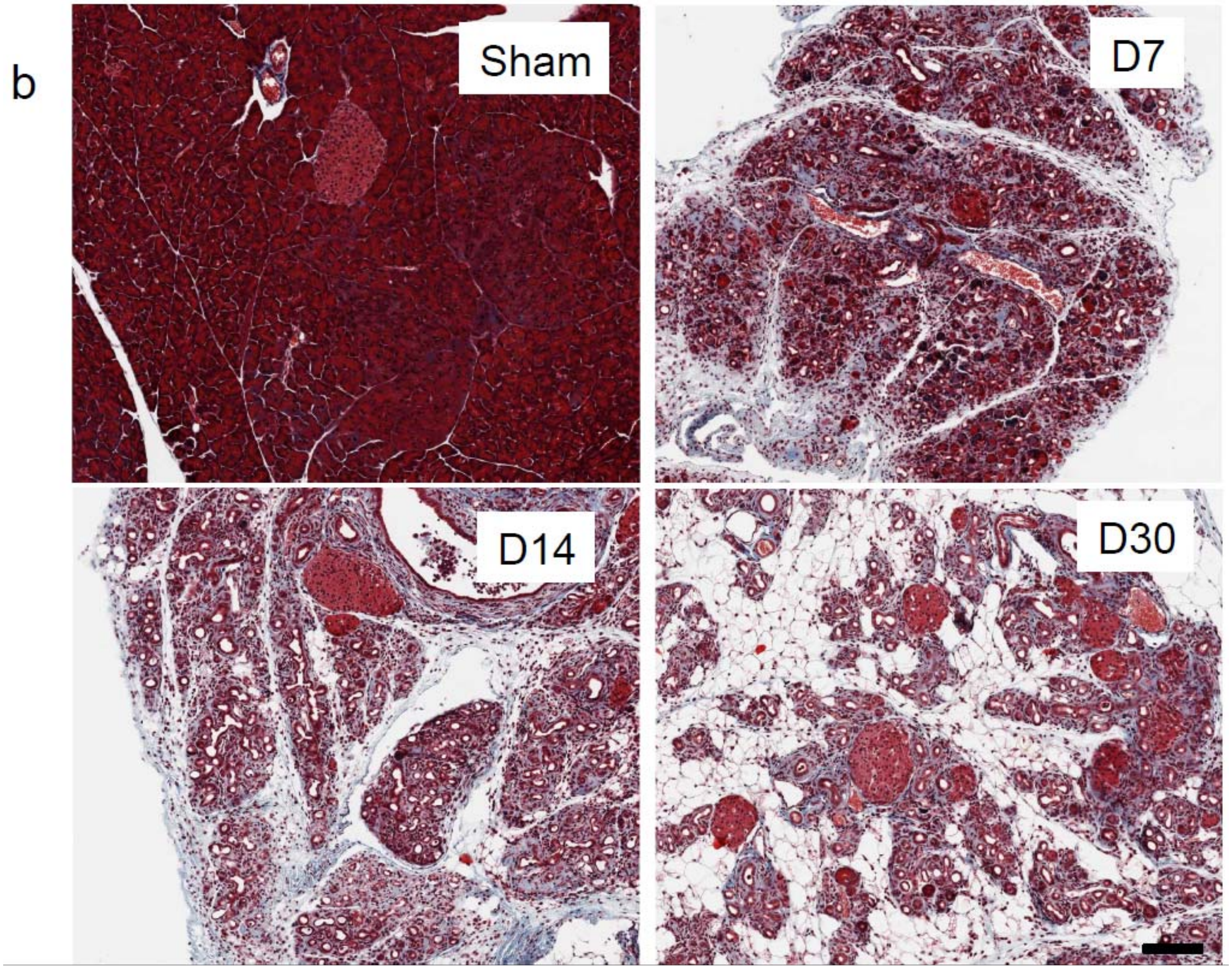
Supplementary Figure 1. Ligation of the splenic lobe of the pancreas does not does not affect body mass or glucose homeostasis in mixed genetic background or Balb/c mice compared to shams; PDL causes major morphologic changes. (a) Ligation suture is placed on the splenic branch of the pancreatic duct. Figure adapted from Page et al, Journal of the Pancreas, 2000, 1(4):191-203. (b-c) Mixed genetic background cohort. Body mass (b) and random fed blood glucose (c). (d-e) Balb/c cohort. Body mass (d) and random fed blood glucose (e). (f-g) Partial pancreatectomy in the Balb/c cohort. Body mass (f) and random fed blood glucose (g). (h-j) Glucose tolerance tests in mixed genetic background cohort at 7 days (h), 14 days (i), and 30 days (j). Mean ± S.E.M., 5-10 animals per group.



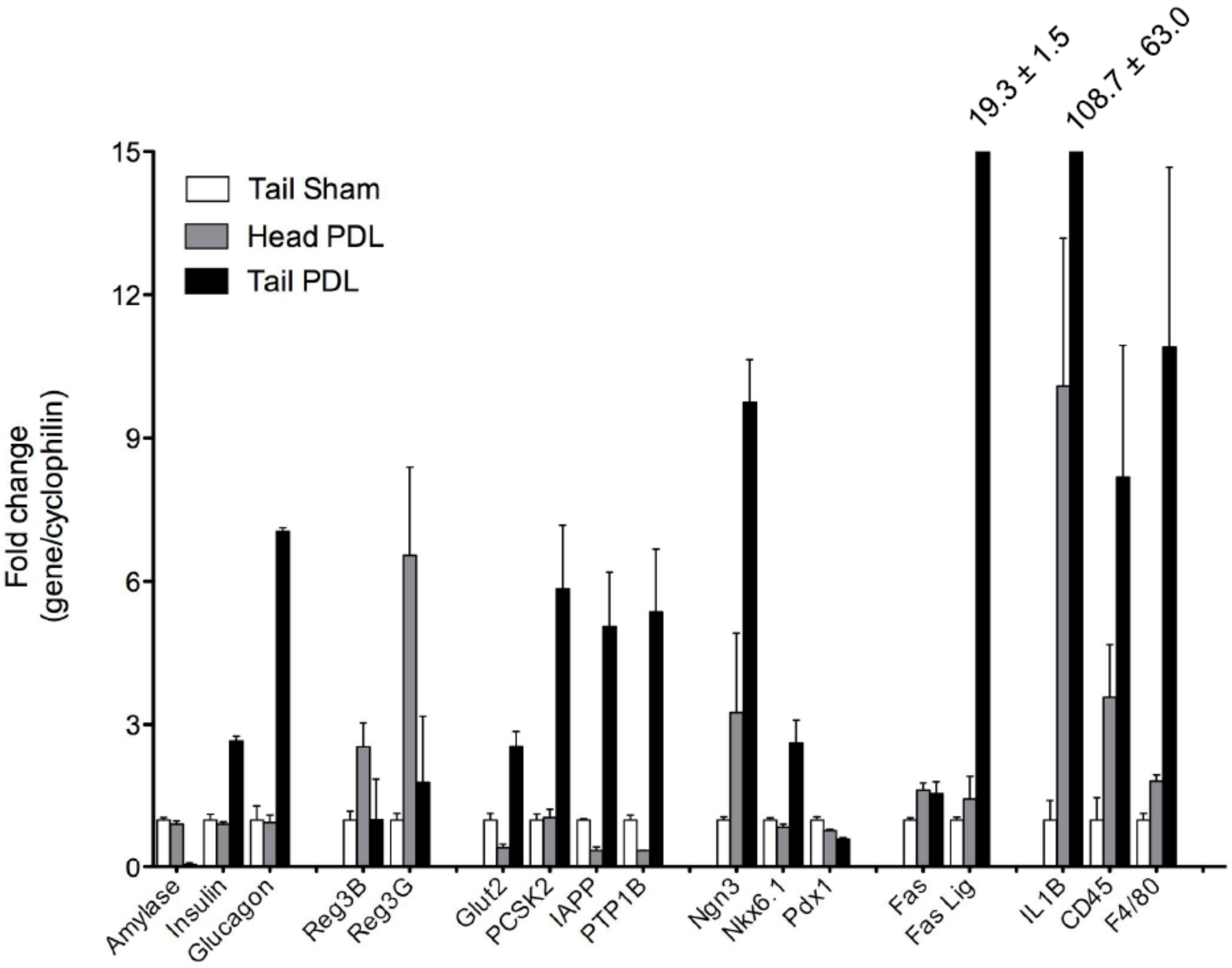

d
Body mass: Balb/c
Pre-Op
Sacrifice
Weight (g)
0
10
20
30
40
50
Sham
PDL
D7
e
Random fed glucose: Balb/c
Sacrifice
Glucose (mg/dl)
0
50
100
150
200
250
Sham
PDL
D7
f
Body mass: Balb/c Parcial pancreatectomy
Pre-Op
Sacrifice
Weight (g)
0
10
20
30
40
50
Sham
PPx
D5
g
Random fed glucose: Balb/c Parcial pancreatectomy
Pre-Op
Sacrifice
Glucose (mg/dl)
0
50
100
150
200
250
Sham
PPx
D5
h
GTT: B6129SF1/J day 7
Sham
PDL
Glucose (mg/dl)
0
100
200
300
400
15
30
60
120
Time (min)
D7
i
GTT: B6129SF1/J day 14
Sham
PDL
Glucose (mg/dl)
0
100
200
300
400
15
30
60
120
Time (min)
D14
j
GTT: B6129SF1/J day 30
Sham
PDL
Glucose (mg/dl)
0
100
200
300
400
15
30
60
120
Time (min)
D30

Supplementary Figure 2. PDL causes major morphologic changes. Characteristic histopathology of DL pancreas stained with hematoxylin and eosin (**a**) and Masson's trichome staining (**b**) of sham and DL tail at 7, 14, and 30 days. Note replacement of acinar tissue by fat and progressive fibrosis. Scale bars: 100 μM.

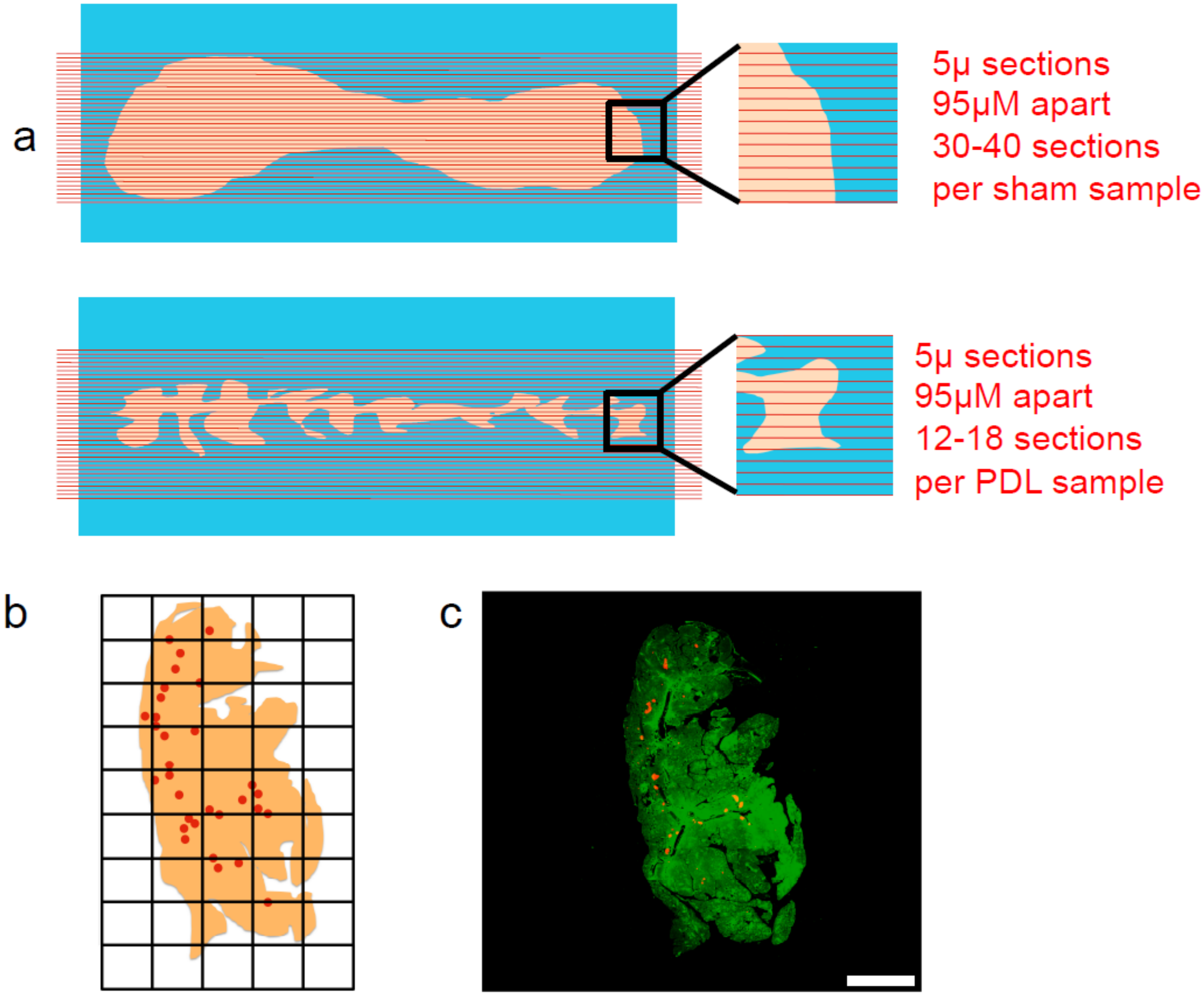


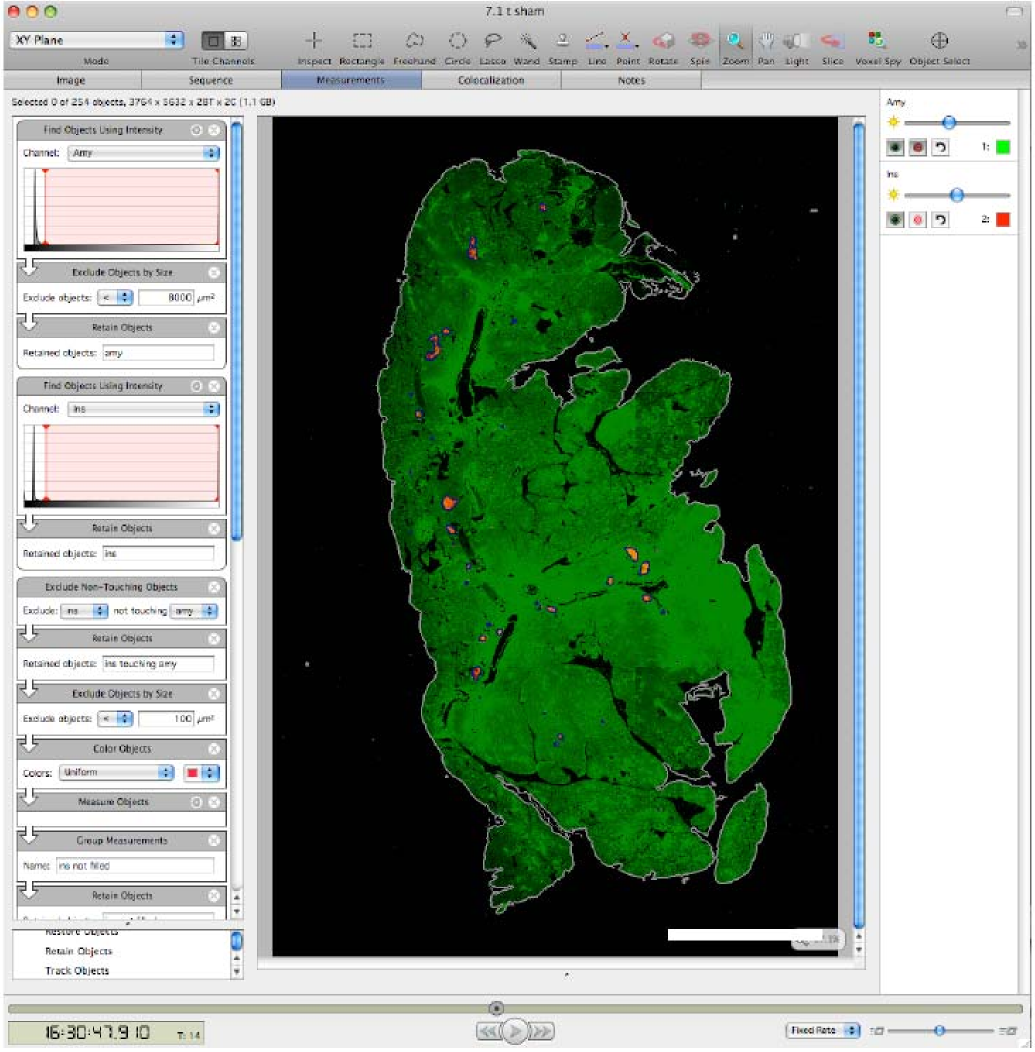

b
Sham
D7
D14
D30

Supplementary Figure 3. qPCR mRNA expression studies of pancreas samples after PDL versus uninjured head or sham-operated control tail. Results expressed as fold change from control when compared to cyclophilin A. Mean ± S.E.M., 3 biological replicates per group.



Supplementary Figure 4. Microscopy and image processing strategy of β-cells. (a) The entire pancreas was sampled at 95µm intervals in the mixed genetic background cohort (160µm intervals were performed in the Balb/c cohort). (b-d) An automated stage was used to scan and tile images from entire longitudinal sections. (b) Schematic. (c) Spliced images, acquired with a 5x objective in the fluorescent channel cy2 (green) for total pancreas and cy3 (yellow) for insulin. (d) Volocity analysis software was used to find the area of total pancreatic tissue (white border) and β-cell tissue (blue border). Scale bars, 2mm.





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Channel: Amy

Exclude Objects by Size
Exclude objects: < 8000 µm²

Retain Objects
Retained objects: amy

Find Objects Using Intensity
Channel: ins

Retain Objects
Retained objects: ins

Exclude Non-Touching Objects
Exclude: ins not touching amy

Retain Objects
Retained objects: ins touching amy

Exclude Objects by Size
Exclude objects: < 100 µm²

Color Objects
Colors: Uniform

Measure Objects

Group Measurements
Name: ins not filled

Retain Objects

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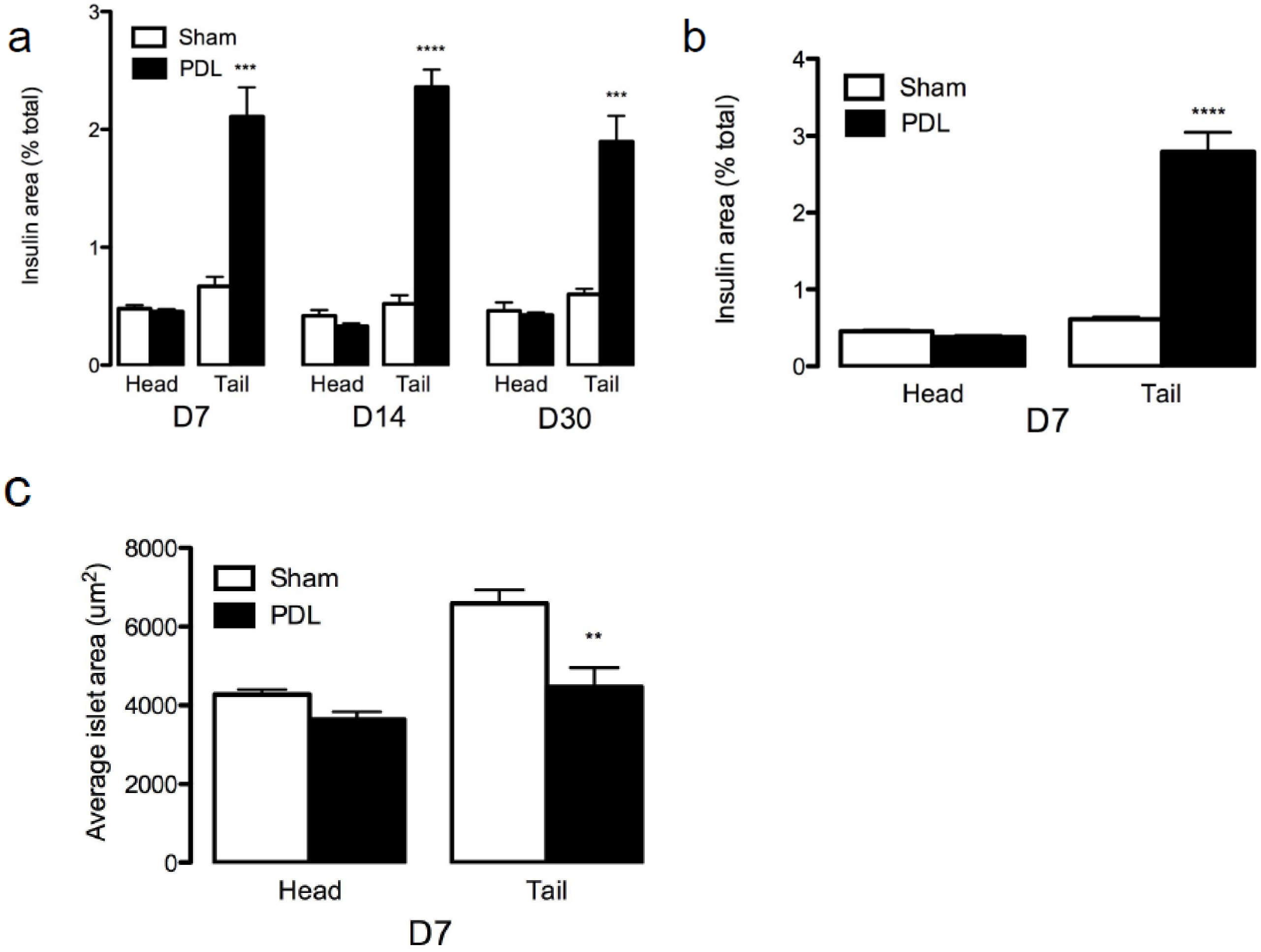
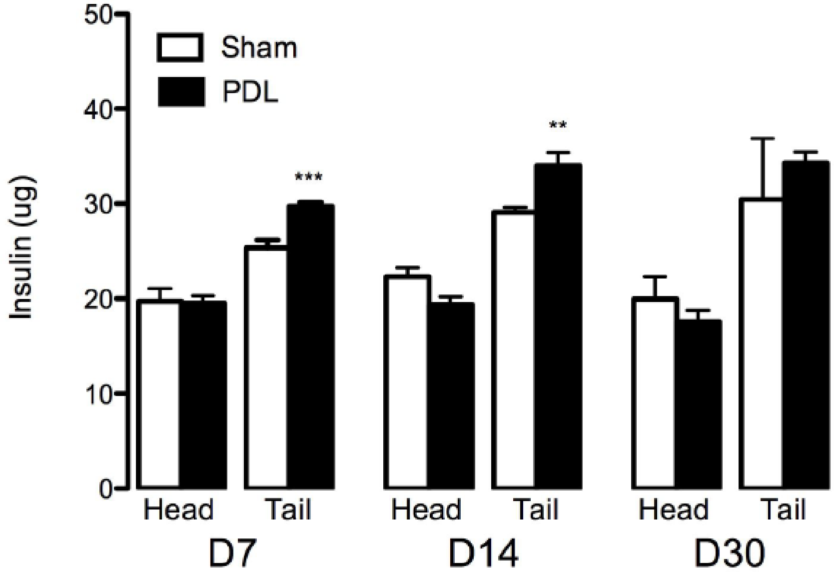
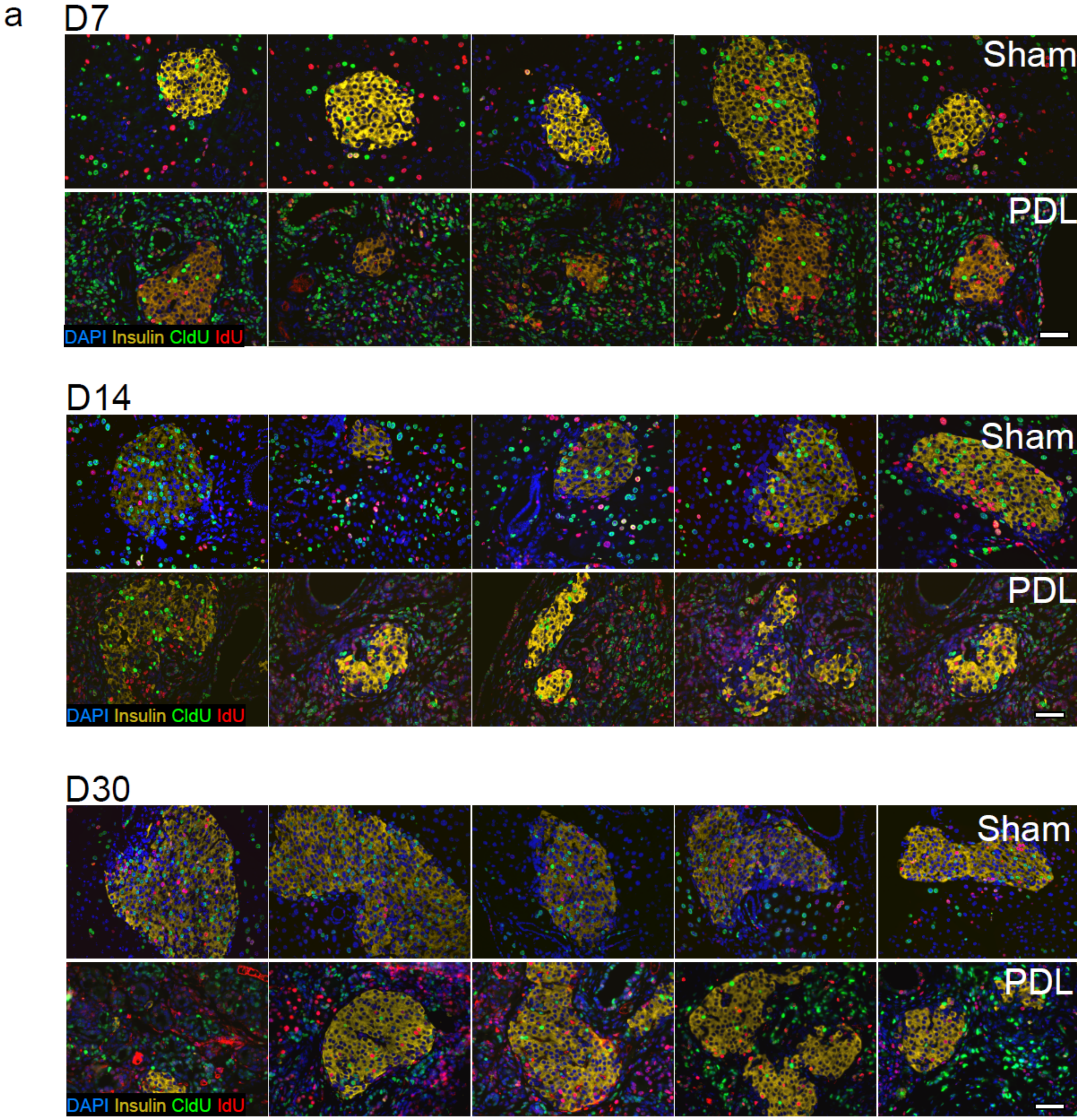
Supplementary Figure 5. PDL increases average insulin area and is associated with reduced islet size (a, b) Quantitative analysis of average β-cell area in mixed genetic background mice (a) and Balb/c mice (b), assessed by insulin$^+$ area compared to total pancreas area. Mean ± S.E.M., 4-5 animals per group. (c) Quantitative analysis of average islet size. Mean ± S.E.M, 4-5 animals per group. **: $p < 0.01$, ***: $p < 0.001$, ****: $p < 0.0001$, ligated vs. unligated pancreas tail.

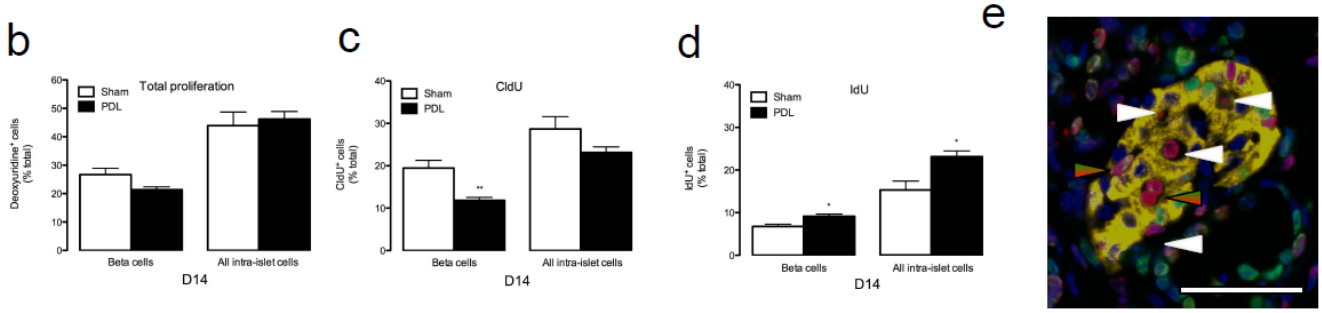


Supplementary Figure 6. Pancreatic insulin content is only slightly increased by PDL. Quantitative analysis of total insulin content from sham or PDL pancreata harvested at day 7. Mean ± S.E.M, 4-5 animals per group. **: $p < 0.01$, ***: $p < 0.001$, ligated vs. unligated pancreas tail.

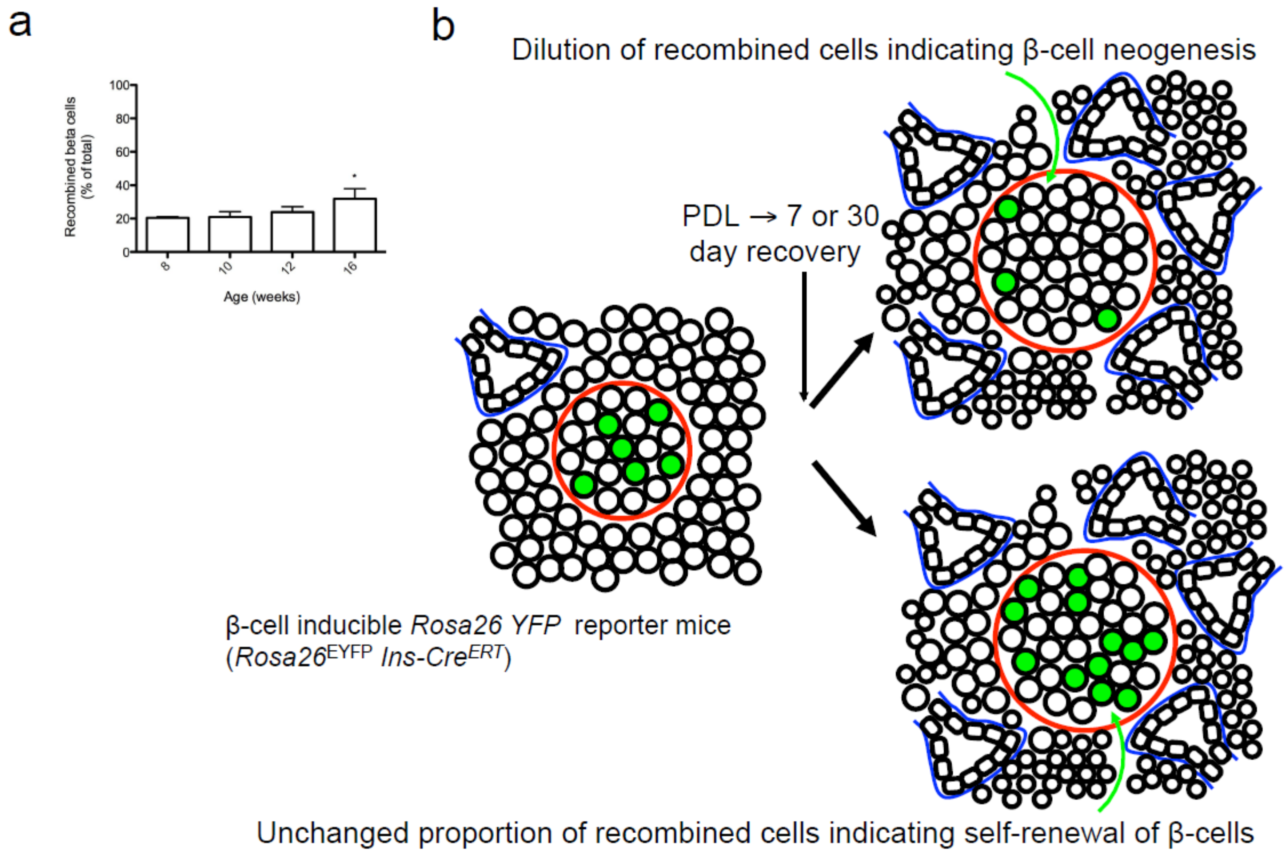


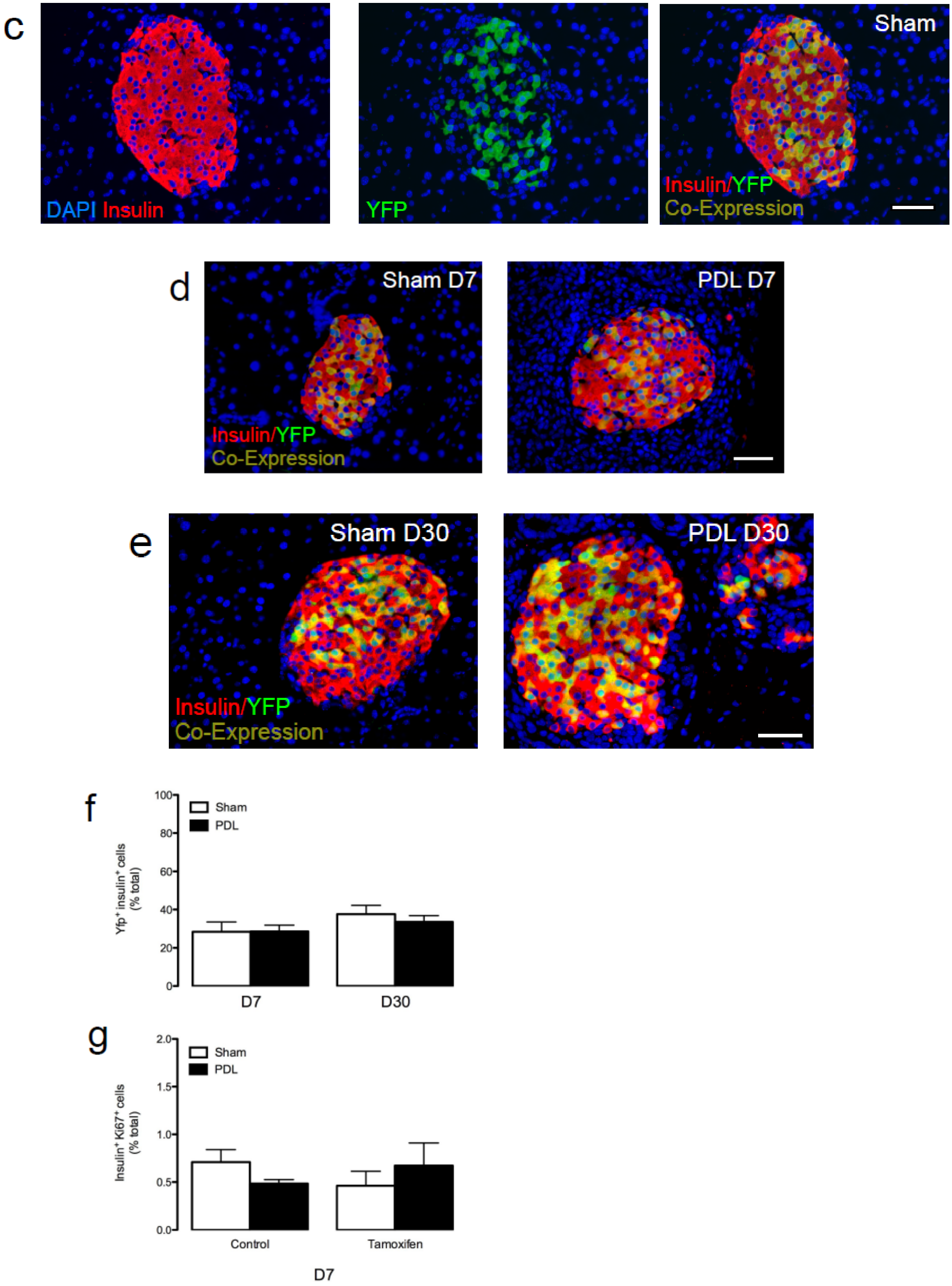
Supplementary Figure 7. PDL does not induce serial proliferation in β-cells or their adult progenitors. (a) Images of islets from sham and PDL tail at 7, 14, and 30 days. DAPI (blue) insulin (yellow), CldU (green), IdU (red). (b-d) Quantitative analysis of thymidine incorporation in β-cells or total intra-islet cells after thymidine analogue labeling, as detected by cells that contained any deoxyuridine analogue (b), CldU$^+$ (c), or IdU$^+$ (d). Mean ± S.E.M., 4-5 animals per group. (e) Representative image of CldU$^+$ and IdU$^+$ intra-islet nuclei after PDL. White triangles denote non- β-cells that were CldU$^+$ or IdU$^+$. Red/green triangles denote non- β-cells that were both CldU$^+$ and IdU$^+$. Scale bars: 50 µM.





Supplementary Figure 8. PDL does not convert non β-cells to dilute the β-cell lineage, as assessed by low-level spontaneous recombination within β-cells. (a) Spontaneous recombination gradually increases between 8 and 16 weeks of age. Quantitative analysis of β-cell recombination in 8,10,12, and 16 week-old mice assessed by the presence of YFP^+ $insulin^+$ cells compared to total $insulin^+$ cells. Mean ± S.E.M., 4-8 animals per group. (b) Genetic lineage tracing scheme used to assess the cellular origin of β-cells after PDL. Experimental design and possible outcomes. (c-e) Spontaneous recombination within β-cells after sham or PDL treatment. Images from vehicle treated sham and PDL pancreas at 7 and 30 days, respectively. DAPI (blue), insulin (red), YFP (green). (f) Quantitative analysis of β-cell recombination, assessed by the presence of YFP^+ $insulin^+$ cells compared to total $insulin^+$ cells. Mean ± S.E.M., 4-5 animals per group. The similar percentages of YFP labeled cells after tamoxifen induction and PDL indicates that β-cells are derived from pre-existing β-cells. (g) Tamoxifen and/or activated CreER had no influence upon β-cell proliferation at 7 days, as assessed by ki67. Mean ± S.E.M., 5 animals per group. Scale bars: 50 µM. *: $p < 0.05$, ligated vs. unligated pancreas tail.




c
DAPI Insulin
YFP
Sham
Insulin/YFP
Co-Expression
d
Sham D7
PDL D7
Insulin/YFP
Co-Expression
e
Sham D30
PDL D30
Insulin/YFP
Co-Expression
f
Yfp+ insulin+ cells (% total)
100
80
60
40
20
0
Sham
PDL
D7
D30
g
Insulin+ Ki67+ cells (% total)
2.0
1.5
1.0
0.5
0.0
Sham
PDL
Control
Tamoxifen
D7

Supplementary Figure 9. Variable recombination in UBC-CreER ROSA reporter mice before and after ductal ligation. Dose dependent recombination in a variety of pancreatic tissues in the basal state within individual mice.