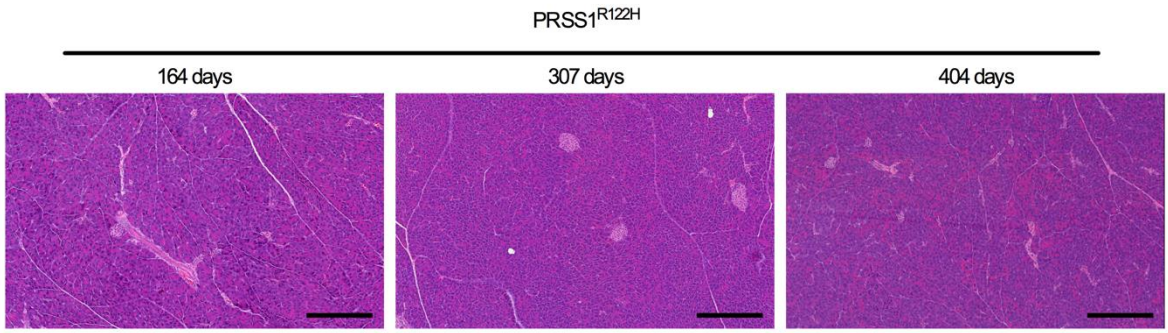
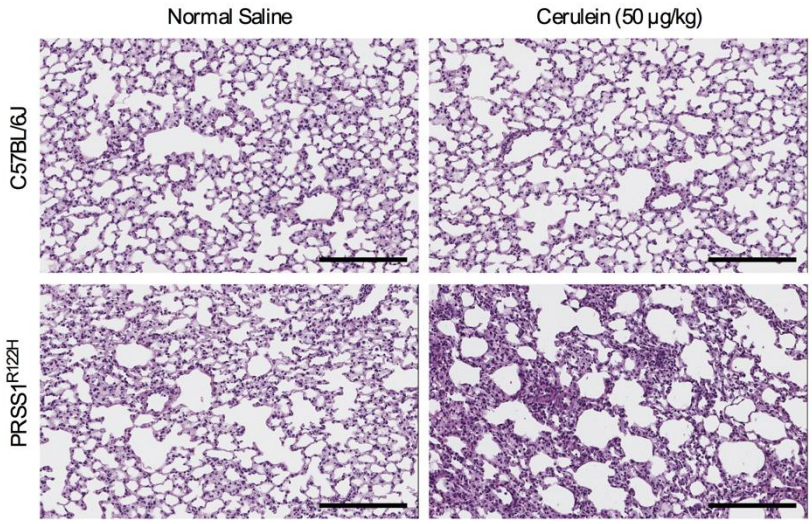
Supplemental Figures



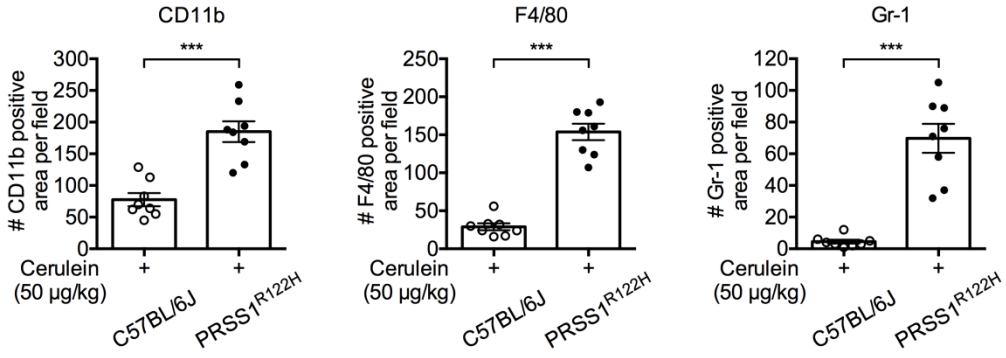
Supplemental Figure 1. No spontaneous pancreatitis was observed in PRSS1^{R122H} mice up to 400 days old.

Pancreata were harvested from PRSS1^{R122H} mice at the indicated ages and stained with Hematoxylin-eosin (Scale bar, 200 μm).

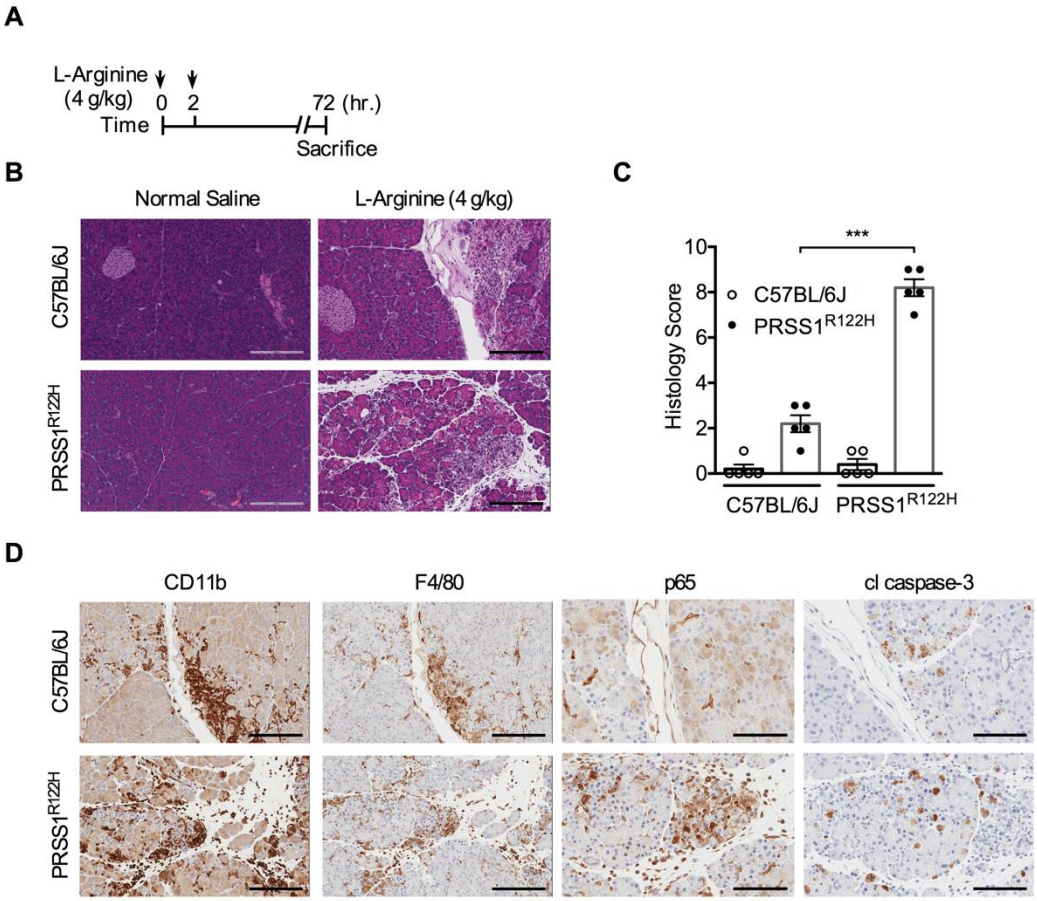


Supplemental Figure 2. Lung inflammation was more severe in PRSS1^{R122H} mice 24 hours after cerulein induction.

Lung was harvested from C57BL/6 and PRSS1^{R122H} mice 24 hours after cerulein-induced acute pancreatitis. Images shown were representative hematoxylin-eosin staining (n = 8). Scale bar, 200 µm.

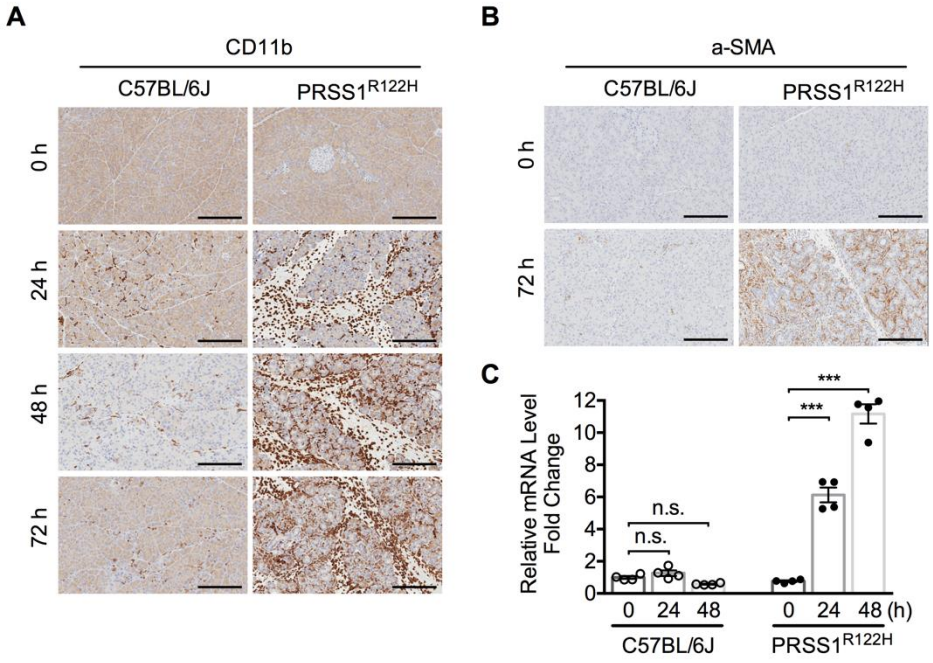


Supplemental Figure 3. Quantification of CD11b, F4/80 and Gr-1 staining in the pancreata of PRSS1^{R122H} mice and C57BL/6J mice. Data were shown as mean ± SEM (n = 8). ****P*<.001; two-tailed unpaired Student's *t*-test.



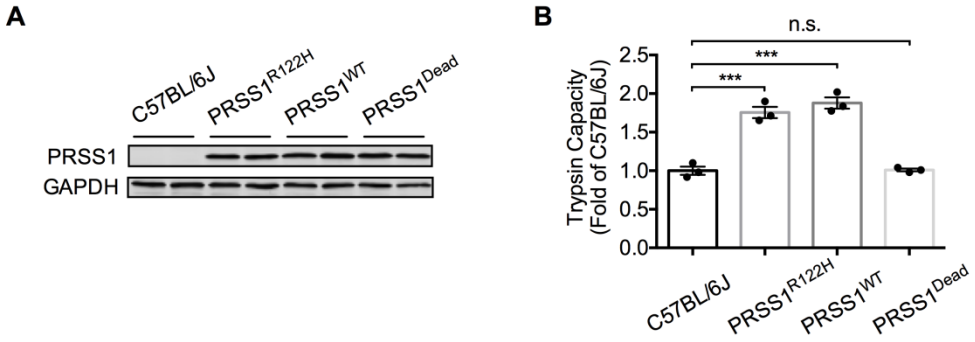
Supplemental Figure 4. PRSS1^{R122H} mice exhibited increased sensitivity to L-arginine-induced pancreatitis.

(A) Schema of L-arginine-induced acute pancreatitis protocol in PRSS1^{R122H} mice and wild-type C57BL/6J mice. **(B)** 72h after L-arginine induction, pancreata were harvested. Hematoxylin-eosin staining was performed (n = 5). Scale bar 200 μm. **(C)** Histology score was shown as mean ± SEM (n=5). ***P<.001 (two-way ANOVA with Tukey's test). **(D)** CD11b, F4/80, p65 nuclear translocation and cleaved caspase-3 expression were detected by immunohistochemical analysis in PRSS1^{R122H} mice and control mice (n=5). Scale bar 200 μm.



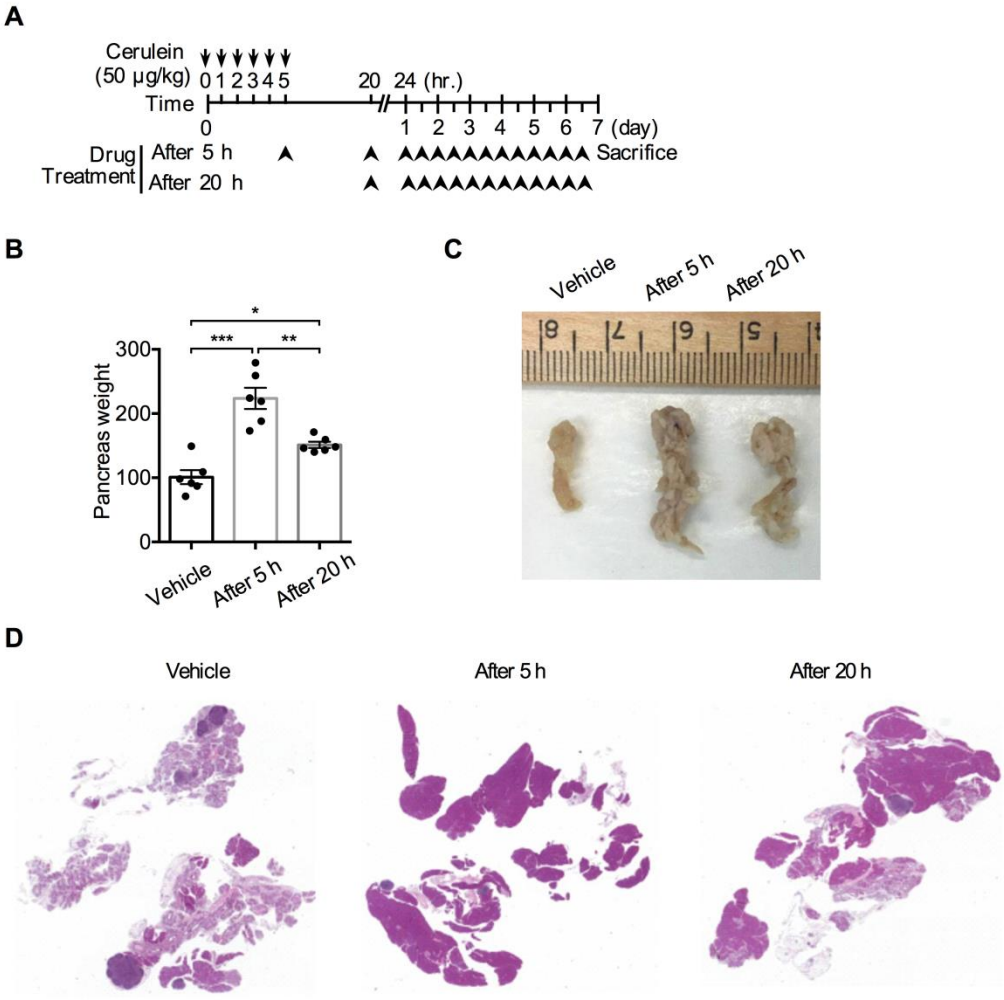
Supplemental Figure 5. Progressive pancreatic damages in PRSS1^{R122H} mice.

(A) Immunohistochemical analysis of CD11b, a marker of leukocytes, showed persistent inflammation in PRSS1^{R122H} mice. In contrast, mild and transient inflammation was observed in similarly treated C57BL/6J mice. Representative of 5 mice per group. Scale bar, 200 μm. **(B)** Positive immunohistochemical staining of a-SMA expression indicated stellate cell activation in PRSS1^{R122H} mice. Representative of 5 mice per group. Scale bar, 200 μm. **(C)** Increased pancreatic α-SMA mRNA expression in transgenic PRSS1^{R122H} mice was detected by real time RT-PCR. Results shown were mean ± SEM (n=4). ****P*<.001, n.s.=not significant (two-way ANOVA with Tukey's test).



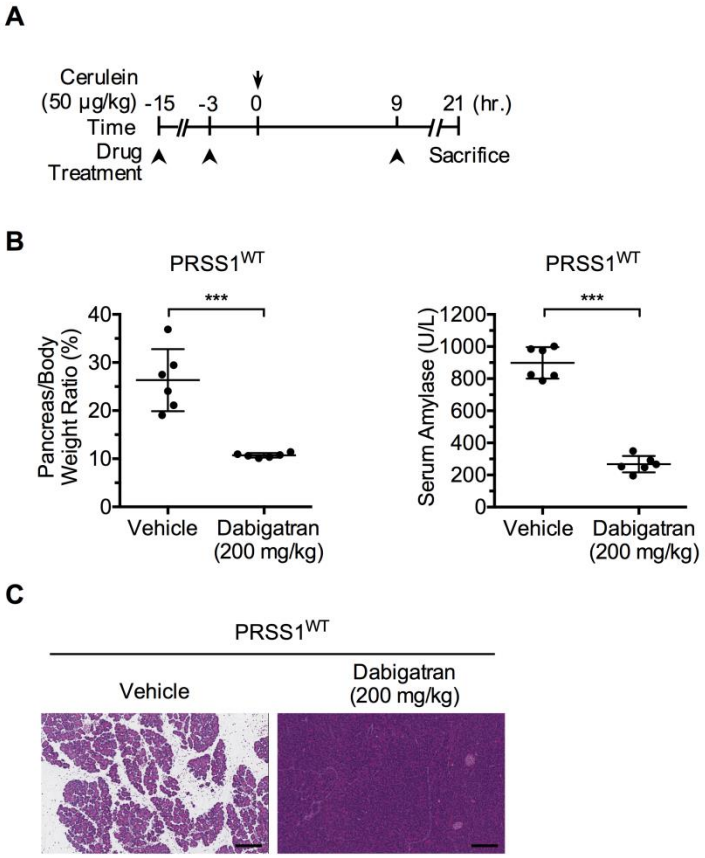
Supplemental Figure 6. Transgenic PRSS1 expression and trypsin capacity in C57BL/6, PRSS1^{R122H}, PRSS1^{WT} and PRSS1^{Dead} mice.

(A) Western blot showed similar expression levels of PRSS1 in these transgenic mice (part of the blot was presented in Fig. 5B). **(B)** Pancreatic lysate from these mice was incubated enteropeptidase to activate trypsinogen. Data were normalized to the level of trypsin capacity in C57BL/6J mice. Higher trypsin capacity was observed in PRSS1^{R122H} and PRSS1^{WT} mice. PRSS1^{Dead} expression did not increase trypsin capacity, suggesting that this mutant PRSS1 is enzymatically inactive. Results were shown as mean ± SEM (n=3). ****P*<.001, n.s., not significant (two-way ANOVA with Tukey's test).

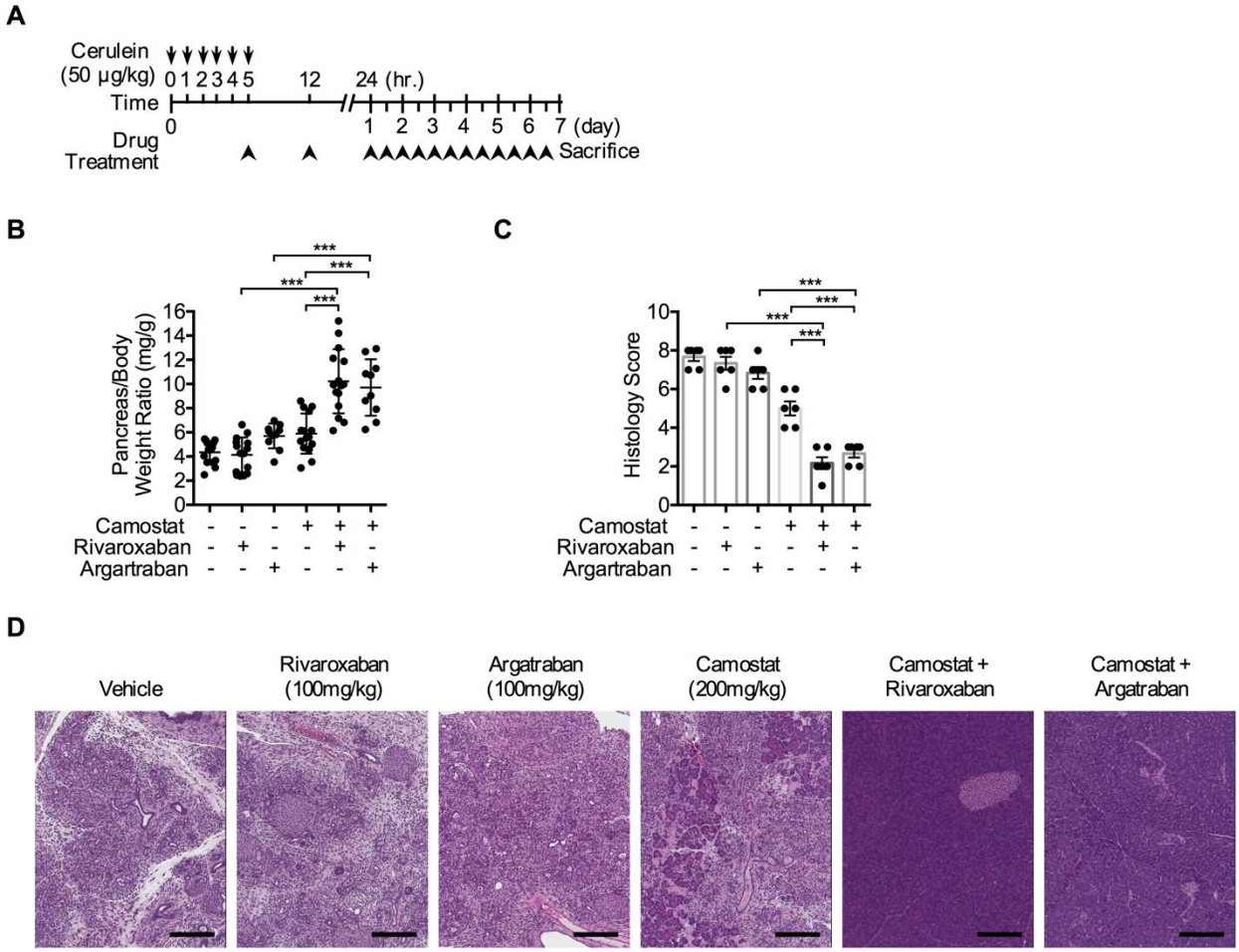


Supplemental Figure 7. Earlier therapy with Dabigatran had better therapeutic efficacy in cerulein-induced pancreatitis.

(A) Treatment with Dabigatran (200 mg/kg) started at 5 hours or 20 hours after cerulein induction. Drug was given twice daily by oral gavage for 7 days. **(B)** Earlier therapy with Dabigatran showed better therapeutic effect, as indicated by pancreas weight preservation. mean ± SEM (n=6). *P<.05, *P<.01, ***P<.001, (one-way ANOVA with Tukey's test). **(C)** Representative images of pancreata after 7 days treatment were shown (n=6). **(D)** Representative images of hematoxylin-eosin (H&E) staining after drug treatment (n=6).



Supplemental Figure 8. Dabigatran prevented the development of AP in mice expressing WT PRSS1. (A) Schema of pancreatitis induction and treatment in PRSS1^{WT} mice. **(B)** Dabigatran prevented the development of AP as indicated by lower pancreatic edema (pancreas-to-body weight ratio, left panel) and serum amylase levels (right panel). mean ± SEM (n = 5 per group). ***P<.001; two-tailed unpaired Student's t-test. **(C)** Representative images of hematoxylin-eosin (H&E) staining after drug treatments (n = 6). Scale bar, 200 µm.



Supplemental Figure 9. Combination of anticoagulation and trypsin inhibition improved the pancreatitis therapy.

(A) Schema of pancreatitis induction and drug treatments with anticoagulation specific agents (Rivaroxaban and Argatroban), trypsin specific inhibitor (Camostat), or in combinations 5h after pancreatitis induction. Drugs were given twice daily by oral gavage over 7 days. Rivaroxaban (Janssen Pharmaceuticals, Inc; *Ki* Factor Xa 0.4nM, trypsin >20,000 nM) is a factor Xa inhibitor, and Argatroban is a thrombin inhibitor (Toronto Research Chemicals; Ki Factor IIa 40 nM, trypsin >20,000 nM) **(B)** Combination of anticoagulation and trypsin inhibition improved pancreatitis therapy, as indicated by pancreas weight preservation. Mean ± SEM (n = 10-15). ***P<.001; one-way ANOVA with Tukey's test. **(C)** Histology score evaluation. Mean ± SEM (n = 6). ***P<.001; one-way ANOVA with

Tukey's test. **(D)** Representative hematoxylin-eosin staining of pancreata from treated mice (n = 10-15). Scale bar, 200 μm.

Supplemental Table 1. q-RT-PCR Primers

Gene Name	Primer Sequence 5'	Primer Sequence 3'
Mcp1	5'- GGTGTCCCAAAGAAGCTG-3'	5'- GTCTGGACCCATTCCTTCT-3'
Il1b	5'-AAATACCTGTGGCCTTGGGC-3'	5'-TTTGGGATCTACACTCTCCAGCT-3'
Tnfa	5'-CCTGTAGCCCACGTCGTA-3'	5'-CCATCGGCTGGCACCACTA-3'
Il6	5'-GTAGCCGCCCCACACAGA-3'	5'-CATGTCTCCTTTCTCAGGGCTG-3'
Atf4	5'-CTTGGCCAGTGCCTCAGACA-3'	5'-CATGGTTTCCAGGTCATCCA-3'
Xbp1s	5'-GGTCTGCTGAGTCCGCAGCAG-3'	5'-CAACTTGTCCAGAATGCCCA-3'
Duox1	5'-CTGCGCTCCATCACCCACT-3'	5'-CAGGGGGACCACAGCTAA-3'
Gpx4	5'-GCATGCTGGGAAATGCCAT-3'	5'-CAGGTCCTTCTCTATCACCT-3'
Rps6	5'-CGCCAGTATGTTGTCAGGAA-3'	5'-GTTGCAGGACACGAGGAGT-3'
Gapdh	5'-CCCCACTTGATTTTGGAGGGA-3'	5'-AGGGCTGCTTTTAACTCTGGT-3'

Abbreviations: *Mcp1*, monocyte chemoattractant protein-1; *IL*, interleukin; *Tnfa,* Tumor Necrosis Factor alpha; *Atf4*, activating transcription factor 4; *Xbp1s,* X-box binding protein 1spliced; *Duox1*, Dual oxidase 1; *Gpx4*, Glutathione peroxidase 4; *Rps6*; ribosomal protein S6; *Gapdh*, glyceraldehyde 3-phosphate dehydrogenase.

Supplemental Table 2. Histology score evaluation

Acute pancreatitis Edema	
Edema	
0	Absent
1	Expansion of interlobular septae
2	Expansion of interlobular septae
3	Expansion of interacinar septae
4	Expansion of intercellular septae
Inflammatory cell	
0	Absent
1	In ducts (around ductal margins)
2	In the parenchyma (< 33% of the lobules)
3	In the parenchyma (> 33% and <67% of the lobules)
4	In the parenchyma (> 67% of the lobules)
Acinar cell death	
0	Absent
1	Focal acinar cell death (< 5%)
2	Focal acinar cell death (5–20%)
3	Diffuse acinar cell death (20–50%)
4	Diffuse acinar cell death (>50%)

Chronic pancreatitis Fibrosis	
Fibrosis	
	0 Absent
	1 Focal (< 5%)
	2 Focal (5–20%)
	3 Diffuse (20–50%)
	4 Diffuse (>50%)
Inflammatory cell	
	0 Absent
	1 In ducts (around ductal margins)
	2 In the parenchyma (< 33% of the lobules)
	3 In the parenchyma (> 33% and <67% of the lobules)
	4 In the parenchyma (> 67% of the lobules)
Fat replacement	
	0 Absent
	1 Focal (< 5%)
	2 Focal (5–20%)
	3 Diffuse (20–50%)
	4 Diffuse (>50%)