Supplemental Figures:

Figure S1

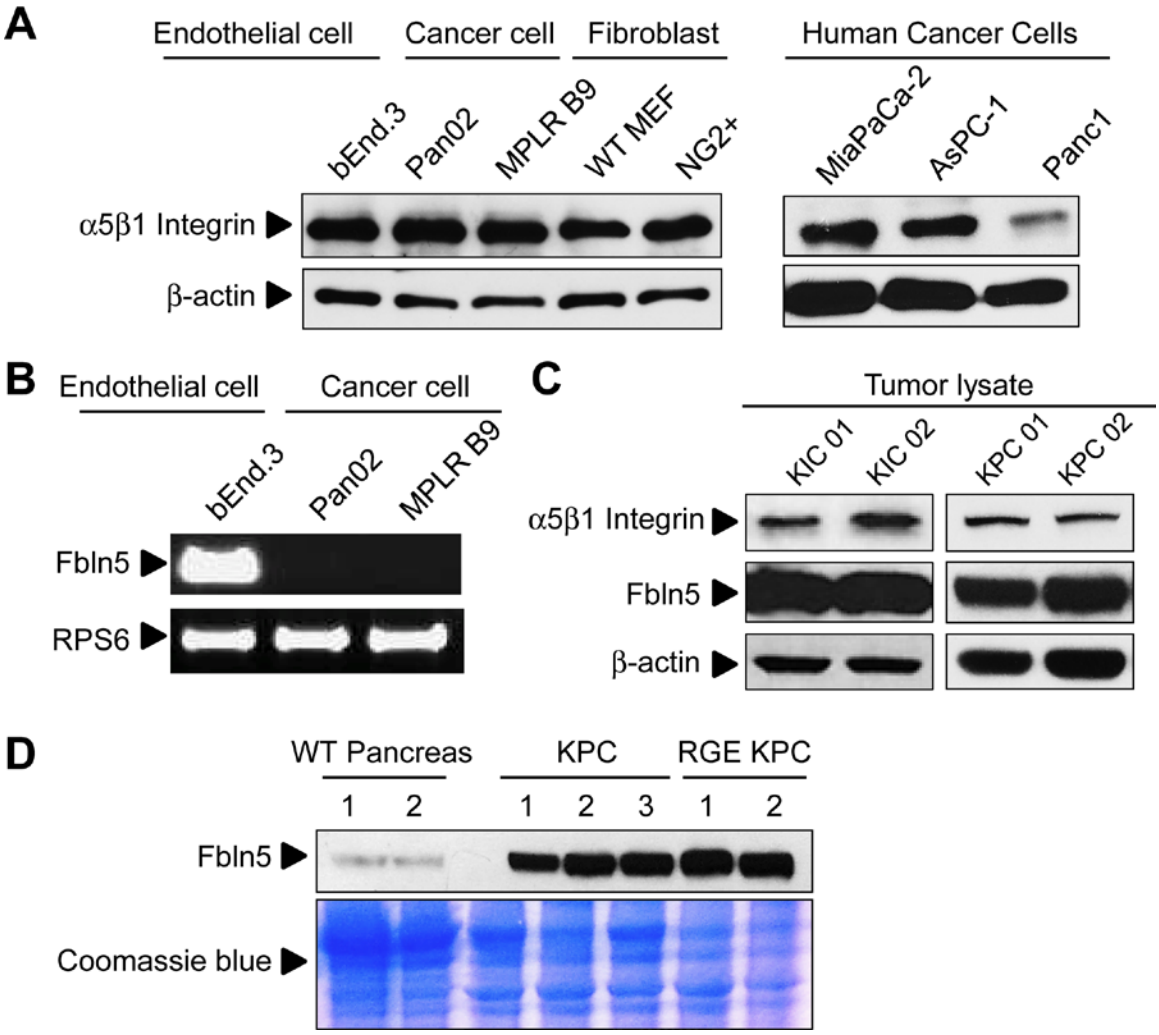

Figure S1. Fbln5 and α5β1 integrin expression pattern in mouse and human. (A) Protein lysates from mouse endothelial, cancer and fibroblast cell lines and human pancreatic cancer cell lines were probed for indicated targets by Western blot. (B) RNA samples from mouse endothelial and cancer cell lines were examined for indicated targets by RT-PCR. (C) Protein lysates from individual mouse (*KIC* or *KPC*) tumor were probed for indicated targets by Western blot. (D) Protein lysates from individual mouse *WT* pancreas, *KPC* and *RGE KPC* tumors were probed for Fbln5 by Western blot. Coomassie blue stained gel was used as loading control.

Figure S2

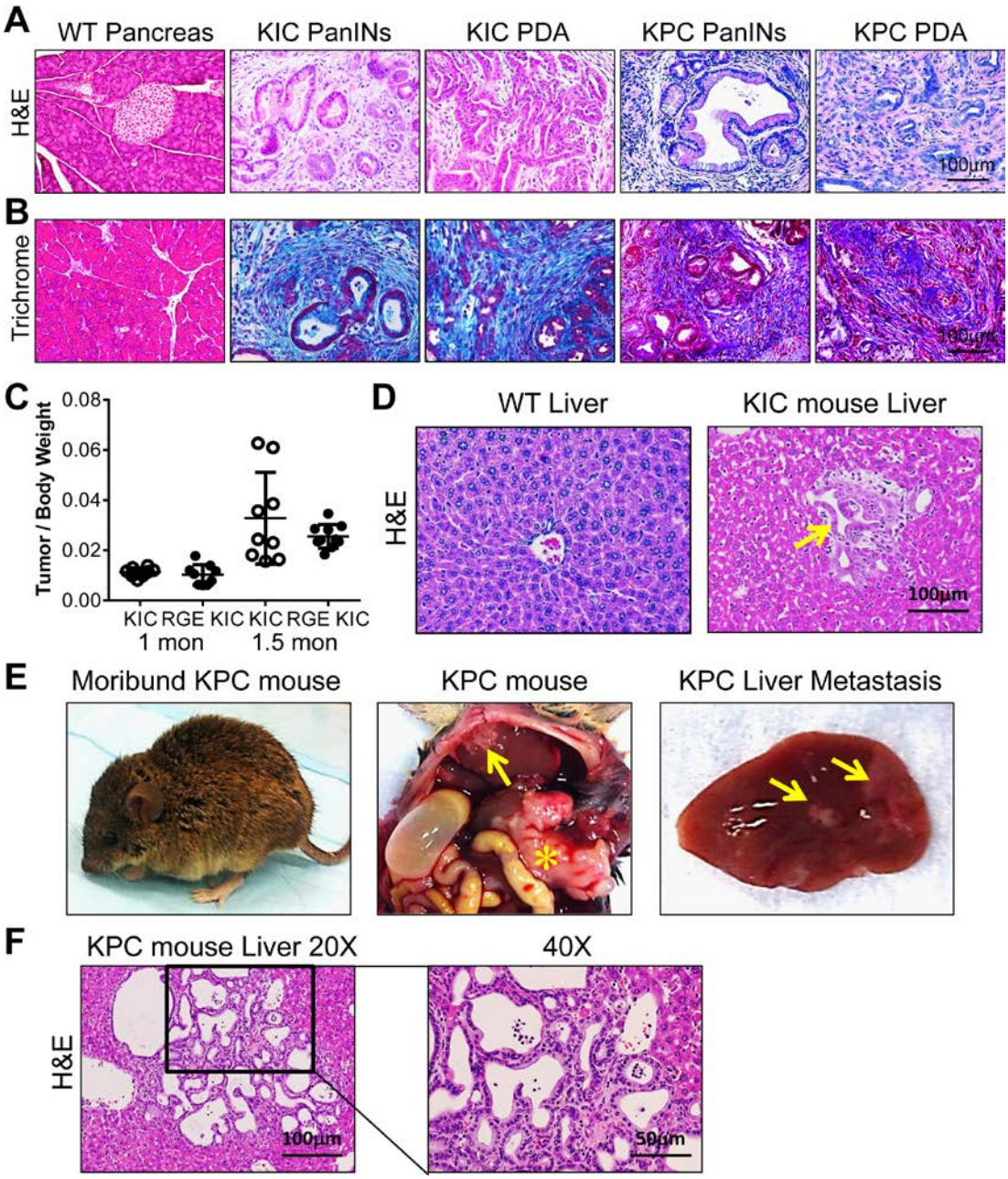

Figure S2. Characterization of *KIC* and *KPC* mice. (A-B) Representative H&E (A) and Masson's trichrome staining (B) of wildtype (*WT*) pancreas, *KIC* and *KPC* tumors. (C) Whole tumors were measured by weights and normalized against body weights from 1 month and 1.5 month (7 week) old *KIC* and *RGE KIC* mice. n≥8 tumors per group. (D) Representative H&E staining of a *KIC* mouse liver after sacrificed for survival study. The liver micrometastasis originating from the primary pancreatic tumor was indicated by a yellow arrow. (E) Representative images of a *KPC* mouse before and after sacrificed for survival study. Primary pancreatic tumor region was indicated by yellow asterisk. Liver metastases were indicated by yellow arrows. (F) Representative H&E staining of a *KPC* mouse liver from the survival study shows liver metastasis. Scale bars are presented as indicated.

Figure S3

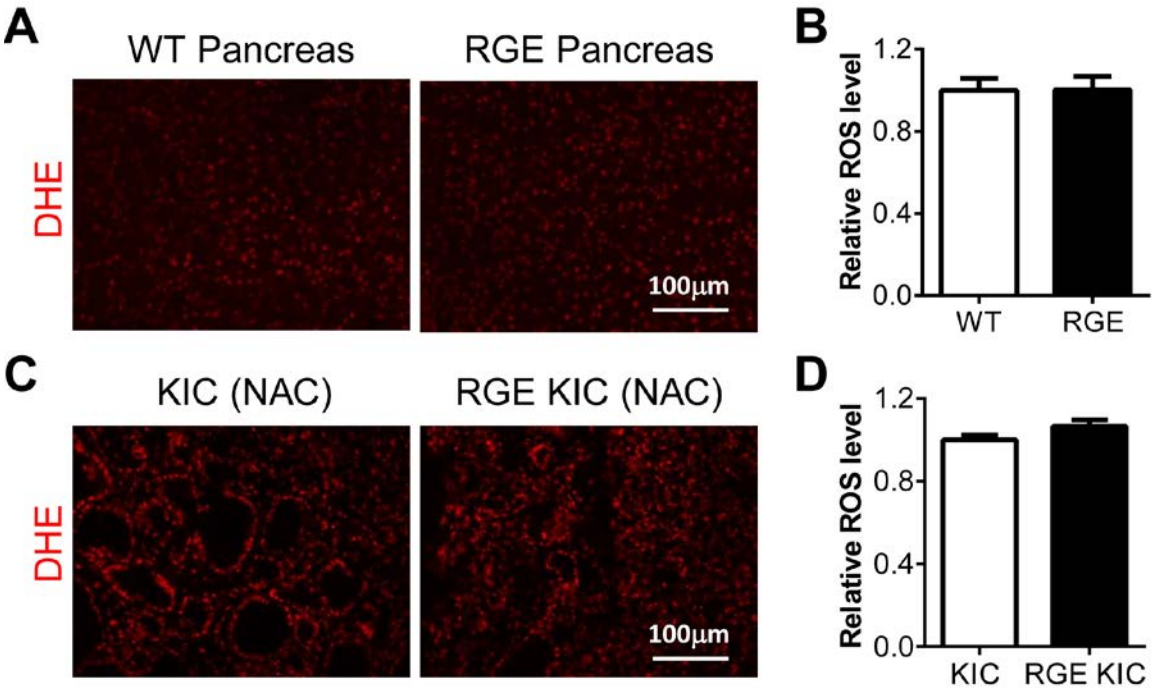

Figure S3. Examination of ROS levels in normal pancreas and NAC-treated *KIC* and *RGE KIC* tumors. (A, C) Dihydroethidium (DHE) (red) staining on freshly cut frozen sections of pancreas from *WT* and *RGE* mice (A), tumors from *KIC* and *RGE KIC* mice treated with antioxidant N-acetyl cysteine (NAC) from 4 week old until moribund (C) for *in situ* detection of ROS. n=3 pancreas and n=4 tumors per group. (B, D) Quantification of DHE staining in panel (A) and (C) were shown in panel (B) and (D), respectively. The relative ROS level was quantified with relative fluorescence intensity measured by the software NIS-Elements. 10 images were taken and quantified per tissue. Scale bars are presented as indicated. All results in (B) and (D) are mean±s.e.m.

Figure S4

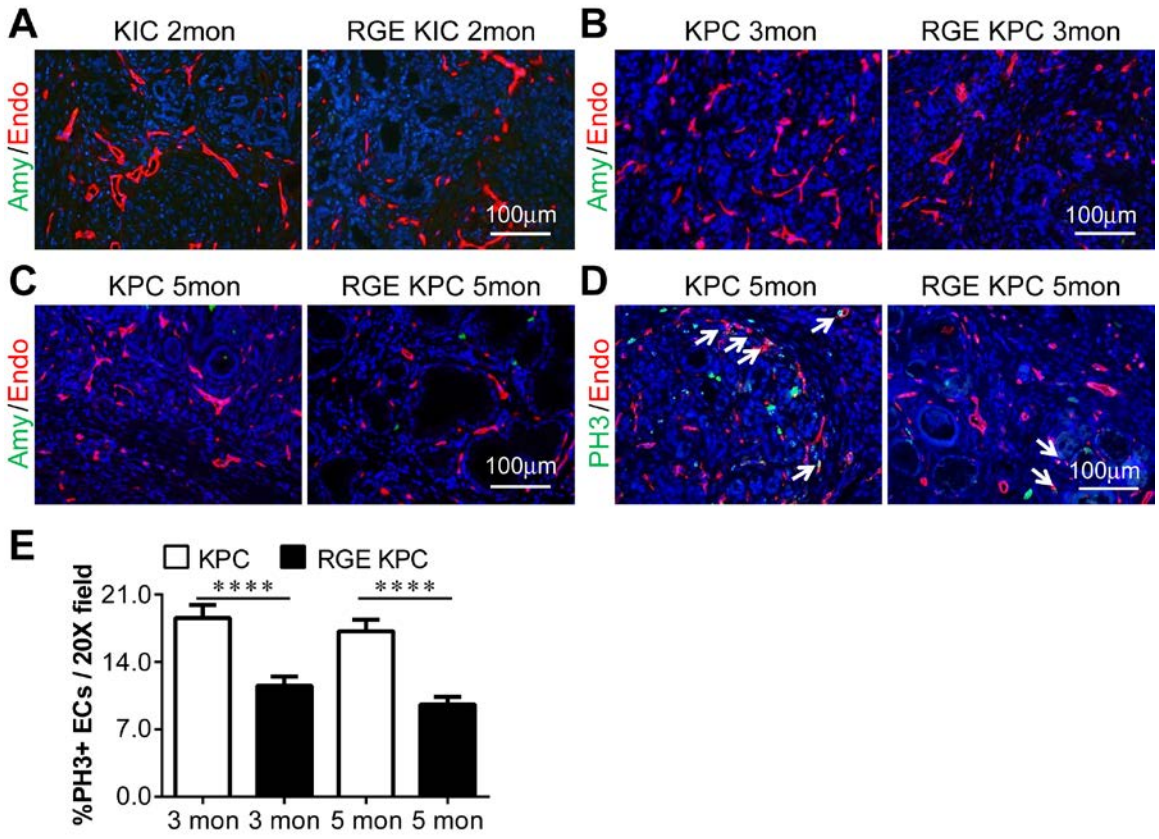

Figure S4. Reduced microvessel density (MVD) in *RGE KIC* and *RGE KPC* tumors compared to *KIC* and *KPC* tumors. (A-C) IF staining on 2 month old *KIC* and *RGE KIC* mouse tumor (A), 3 month (B) and 5 month (C) old *KPC* and *RGE KPC* tumor sections for Amy (green) and Endo (red). n=4-5 tumors per group. (D) IF staining on 5 month *KPC* and *RGE KPC* tumor sections for phospho-Histone H3 (PH3) (green) and Endo (red). Arrows indicate double-labeled ECs. n=4 tumors per group. (E) Quantification of % PH3+ ECs over total number of ECs in 20X field for tumors from 3 and 5 month old *KPC* and *RGE KPC* mice. Results are shown as mean±s.e.m. Unpaired t test was used for statistical analysis. ****, $p<0.0001$. Scale bars are presented as indicated.

Figure S5

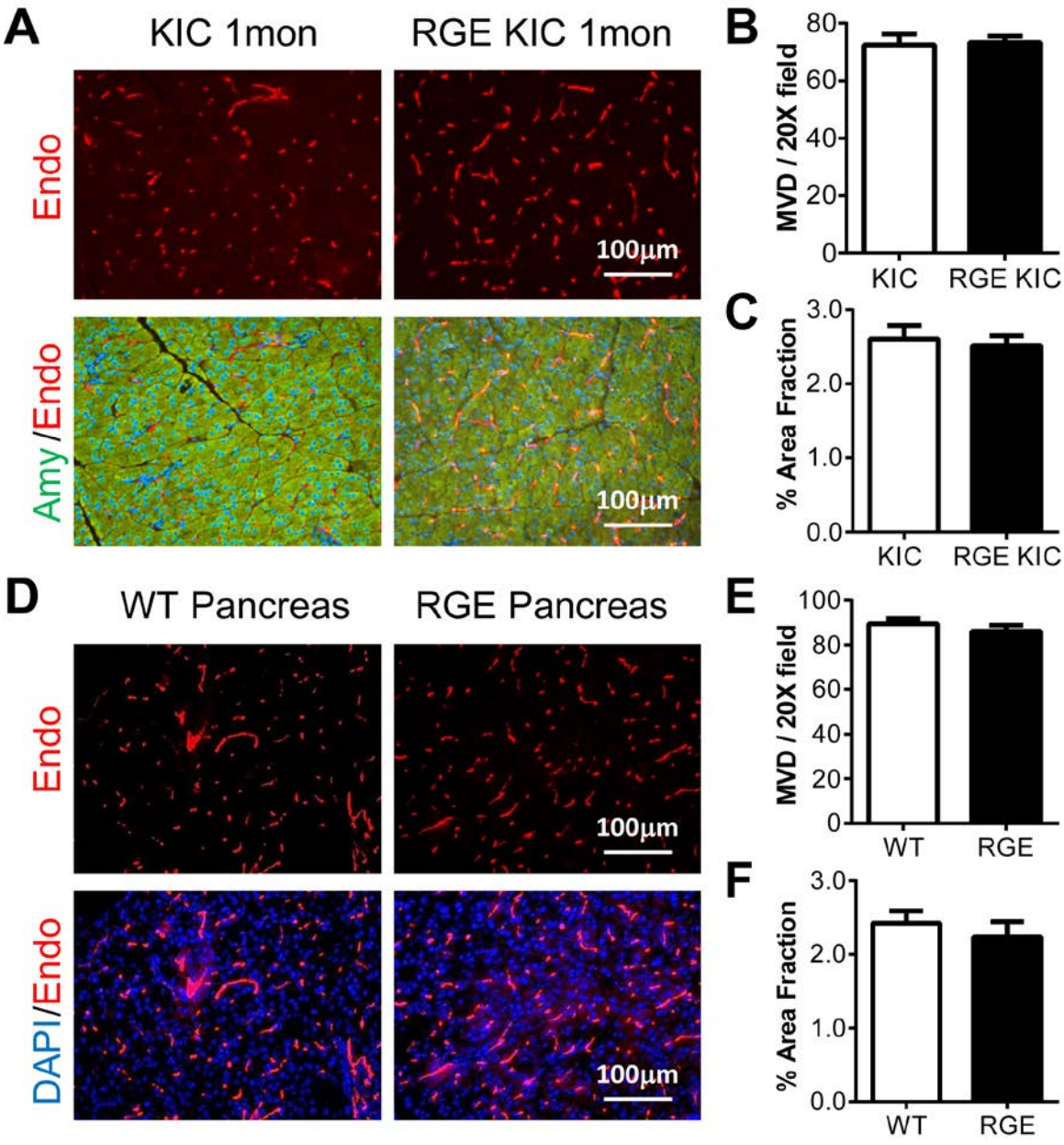

Figure S5. Examination and quantification of MVD in pancreatic tissues from *KIC* and *RGE KIC*, *WT* and *RGE* mice. (A) IF staining on 1 month *KIC* and *RGE KIC* pancreatic tissue sections for Amylase (Amy) (green) and Endomucin (Endo) (red). n=5 tumors per group. (D) IF staining on pancreas sections from *WT* or *RGE* mice for Endo (red). n=3 mice per group. (B, E) MVD was counted per 20X field from 5 tumors (B) or 3 pancreases (E) in each group with 8-10 pictures from each tissue. (C, F) Quantification of MVD for panel (A) or (D) using the software NIS-Elements. Endo stained areas are counted as % area fraction. Scale bars are presented as indicated. All data in (B), (C), (E) and (F) are shown as mean ±s.e.m.

Figure S6

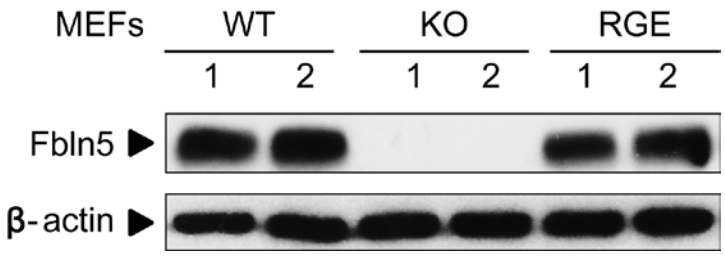

Figure S6. Fbln5 expression in *Fbln5 WT*, *KO* and *RGE* MEFs. Protein lysates from *Fbln5 WT*, *KO* and *RGE* MEFs were probed for indicated targets by Western blot.

Figure S7

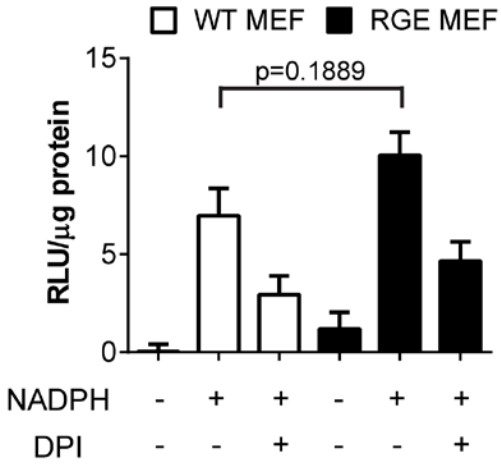

Figure S7. NADPH oxidase (Nox) is not the source of ROS induction for *RGE* MEFs when plated on FN. Primary *WT* and *RGE* MEFs were grown on 10 μg/ml FN-coated plates for 4 hours (hr) with serum free medium (SFM) supplemented with 10 μg/ml FN. Cells were then harvested for Nox activity assay. NADPH serves as substrate. Diphenylene iodonium (DPI) was used as a Nox inhibitor. The relative light unit (RLU) was monitored by a plate reader and the measurement in linear phage (~3 min) was used for quantification. The results are shown as RLU/μg protein. Unpaired t test was used for statistical analysis.