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Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see Authors & Referees and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒ A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐ A description of all covariates tested
☐	☒ A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒ A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒ For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐ For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐ For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐ Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Flow cytometry data acquired using BD FACSDiva software v8.0; Immunofluorescence data captured using Leica Application Suite AF; Immunohistochemistry data captured using Leica LAS 4.2.
Data analysis	Fluorescent/IHC images were analyzed by Leica LAS AF, ImageJ (2.1.0), and Adobe Photoshop (CS6). Statistics and plots were generated by ggplot2 (3.2.1) in R Studio (1.2.5019) and GraphPad Prism (9.1). Flowjo (8.7) was used for FACS data. Reads were paired using fastq-join tool at Galaxy (usegalaxy.org, version 1.1.2-806.1). KRAS variants were identified using custom python scripts (https://github.com/cmroake/KRAS_scripts)

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors/reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

Sequencing data for KRAS variants is available with the SRA accession: PRJNA598774. All other data are available from the authors upon reasonable request.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Sample sizes were set using power calculation, based on effect size, significant level and power level.
Data exclusions	No individual samples or animals were excluded from any analyses.
Replication	All replication experiments were statistically analyzed and presented in the paper.
Randomization	The animal were randomly assigned to each group (control/experimental).
Blinding	The investigators were not blinded to the identities of the samples. All experimental and corresponding control samples were collected and analyzed at the same time under the same condition. Quantification was made with the same software settings.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology
☐	☒ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used

The following antibodies were used in this study: anti-CK19(Hybridoma Bank, TROMA-III, 1:100-1:500); anti-RFP(Abcam, ab124754, 1:500-1:1000); anti-RFP (MBL, M208-3, 1:200); anti- Ki-67 (ThermoFisher, RM-9106-S1, 1:100-1:500); anti BrdU (AbD Serotec, MCA2060, clone 1G9,3G5 (mixed), 1:250-1:500); anti-Amylase (Sigma, A8273, 1:250-1:1000); anti-Sox9 (Millipore, AB5535, clone 2B10, 1:100-1:500); Phospho-p44/42 MAPK (Erk1/2) (Cell signaling, 4370, clone D13.14.4E, 1:250-1:1000); Phospho-p44/42 MAPK (Erk1/2) (Cell signaling, 4376, clone 20G11, 1:250-1:1000); anti-Insulin (Sigma, 273A, 1:250-1-1000); anti-Muc5A (ThermoFisher, MS-145-P0, clone 45M1 , 1:500); anti-DCLK1 (Abcam, ab31704, 1:250); anti-BMI1 (Bethyl, A301-694A, 1:250-1:500); anti-CD45 (Biolegend, 103116, clone 30-F11 , 0.25ug/1mio cells)
Secondary antibodies used were all purchased from Jackson ImmunoResearch and used at 1:500 dilution. Peroxidase AffiniPure Goat Anti-Mouse IgG, light chain specific (#115-035-174), Alexa Fluor® 488 AffiniPure Donkey Anti-Guinea Pig IgG (H+L) (#706-545-148), Peroxidase AffiniPure Donkey Anti-Rat IgG (H+L) (#712-035-153), Alexa Fluor® 488 AffiniPure Donkey Anti-Rat IgG (H+L) (#712-545-153), Alexa Fluor® 488 AffiniPure Donkey Anti-Rat IgG (H+L) (#712-545-153), Cy™3 AffiniPure Donkey Anti-Rabbit IgG (H+L) (#711-165-152) and Peroxidase AffiniPure Donkey Anti-Rabbit IgG (H+L) (#711-035-152).

Validation

anti-CD45 (Biolegend, 103116) was quality control tested by immunofluorescent staining with flow cytometric analysis by the company. The following antibodies were validated for immunoblot and IF of RFP proteins by the manufacturer using gene transfection: anti-RFP(Abcam, ab124754); anti-RFP (MBL, M208-3).
The following antibodies were validated for Immunoblot or Immunohistology of human and mouse proteins by the manufacturer using gene knockout, gene knockdown, transfection, or other biological manipulations: anti-Sox9 (Millipore, AB5535), Phospho-p44/42 MAPK (Erk1/2) (Cell signaling, 4370), Phospho-p44/42 MAPK (Erk1/2) (Cell signaling, 4376), anti-DCLK1 (Abcam, ab31704), anti-BMI1 (Bethyl, A301-694A), anti- Ki-67 (ThermoFisher, RM-9106-S1); anti BrdU (AbD Serotec, MCA2060).
anti-Insulin (Sigma, 273A), anti-CK19(Hybridoma Bank, TROMA-III), anti-Muc5A (ThermoFisher, MS-145-P0) and anti-Amylase (Sigma, A8273) antibodies were validated by Immunohistology in this study.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	Tert CreERT2/+ mice were generated in JM8/F6 mouse ES cells. Ptf1CreERTM/+, Kras-LSL-G12D/+, (Gt(ROSA)26Sortm14(CAG-tdTomato)Hze/J) mice were obtained from the Jackson Laboratory. 8-10 week old mice of mixed background were used for all mouse studies. Both genders were used in this study. Mice were housed in 22°C ambient temperature and 40% humidity, with a 12-hour light/dark cycle (7am – 7pm).
Wild animals	No wild animals were used in this study.
Field-collected samples	No field collected samples were used in the study
Ethics oversight	All animal experiments were approved by the Administrative Panel on Laboratory Animal Care (APLAC) in protocol APLAC-12684.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Human research participants

Policy information about studies involving human research participants

Population characteristics	Individuals having undergone pancreatic resections for neoplastic and non-neoplastic conditions as well as autopsy samples from non pancreatic disease specimens were included in this retrospective study of archival tissue. The age range of the specimens was 24-84 years old and there were 24 female and 22 male specimens analyzed in the study. There were no exclusion criteria related to patient age, gender, or race/ethnicity. Given the fact that pERK+ vs pERK- staining within the specimen is the independent variable, clinical and demographic features do not represent covariates in this analysis
Recruitment	There were no exclusion criteria related to patient age, gender, or race/ethnicity. Cases were included in this retrospective study only on the basis of the presence or absence of histopathologic features relevant to the study design, as described in the Materials/Methods.
Ethics oversight	This study protocol was approved by the Stanford University Institutional Review Board (Protocol #42887)

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	To obtain single cell suspension mice were sacrificed, pancreata harvested and immediately placed in 7ml digestion solution (per pancreas) containing 2 mg/ml Collagenase VIII (Sigma) and 0.2 mg/ml Trypsin inhibitor (Sigma). Pancreata were incubated for 10-12 minutes in a shaker at 37°C. Cells were resuspended with FACS buffer (2% FBS in PBS) and filtered through a 100µm cell strainer. Red blood cells were removed using red blood cell lysis buffer (Ebioscience) for 10 min at RT. Cells were incubated with anti-CD45 antibody (Biolegend) for 45min. Cells were sorted with a BD Aria II flow cytometer using a 100 µm nozzle. Dead cells were excluded based on DAPI (1µM) incorporation. FACS data was analyzed using Flowjo (8.7).
Instrument	FACSAria 2
Software	BD FACSDiva software v8.0 and FlowJo (8.7).
Cell population abundance	Approximately 0.1-1% of total acinar cells were Tomato+.
Gating strategy	Cells were selected by scatter properties. Single cells were gated by the area and width of forward scatter. Viable cells were selected by DAPI exclusion. Acinar cells were selected by CD45 exclusion. Tomato+ and Tomato- acinar cells were selected by Tomato expression compared to control sample.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.