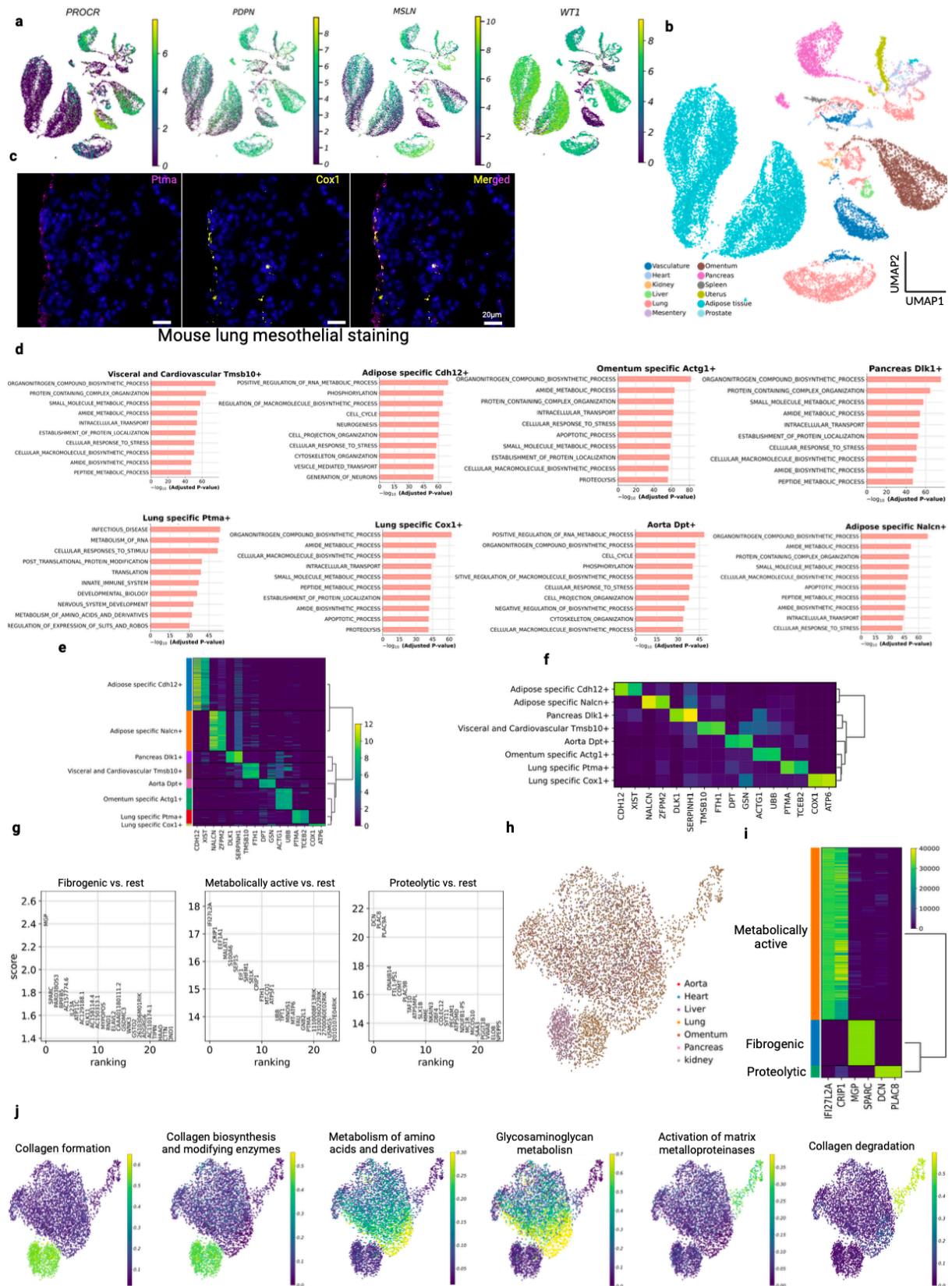
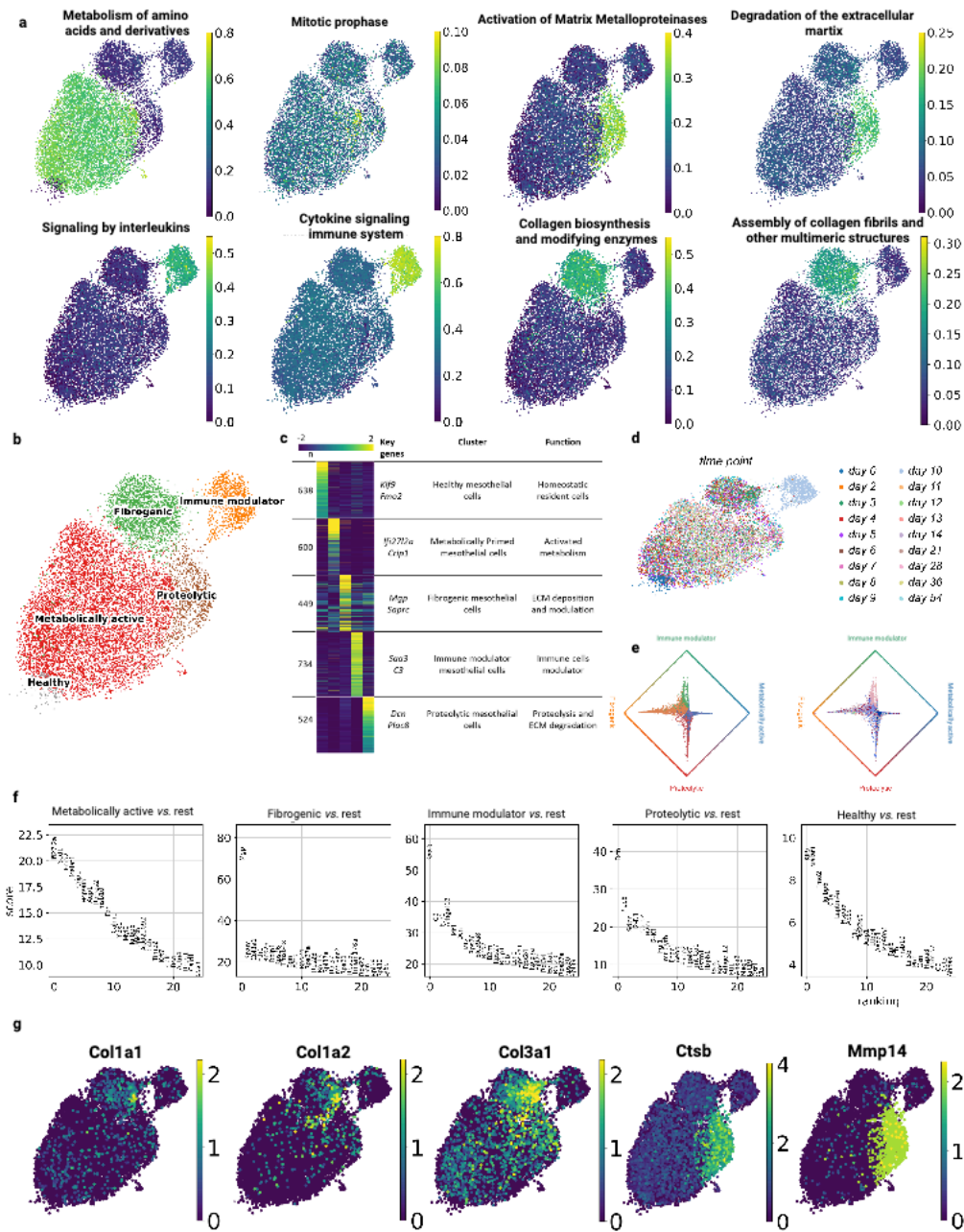




Supplementary Fig. 1: Mouse mesothelial cell steady-state and disease atlas

- a. UMAP embedding of the steady-state mouse mesothelial cell atlas color coded by tissue of origin.
- b. UMAP highlighting the expression levels of the positive used markers for mesothelial cells selection.
- c. Healthy mouse lung immunostaining with Cox1 and Ptma. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm
- d. Pathway enrichment analysis of the different mesothelial cell clusters in the steady-state atlas.
- e. Heatmap of the top two gene markers of each cluster in the steady-state atlas.
- f. Heatmap of the differentially expressed genes between the different clusters in steady-state mouse atlas.
- g. Top twenty-five gene markers of the three identified mouse mesothelial disease-state clusters.
- h. UMAP embedding of the disease-state mouse mesothelial cell atlas color coded by tissue of origin.
- i. Heatmap of the differentially expressed genes between the different clusters in mouse disease-state.
- j. UMAP of the expression level of enriched reactomes in the different clusters at cellular level.

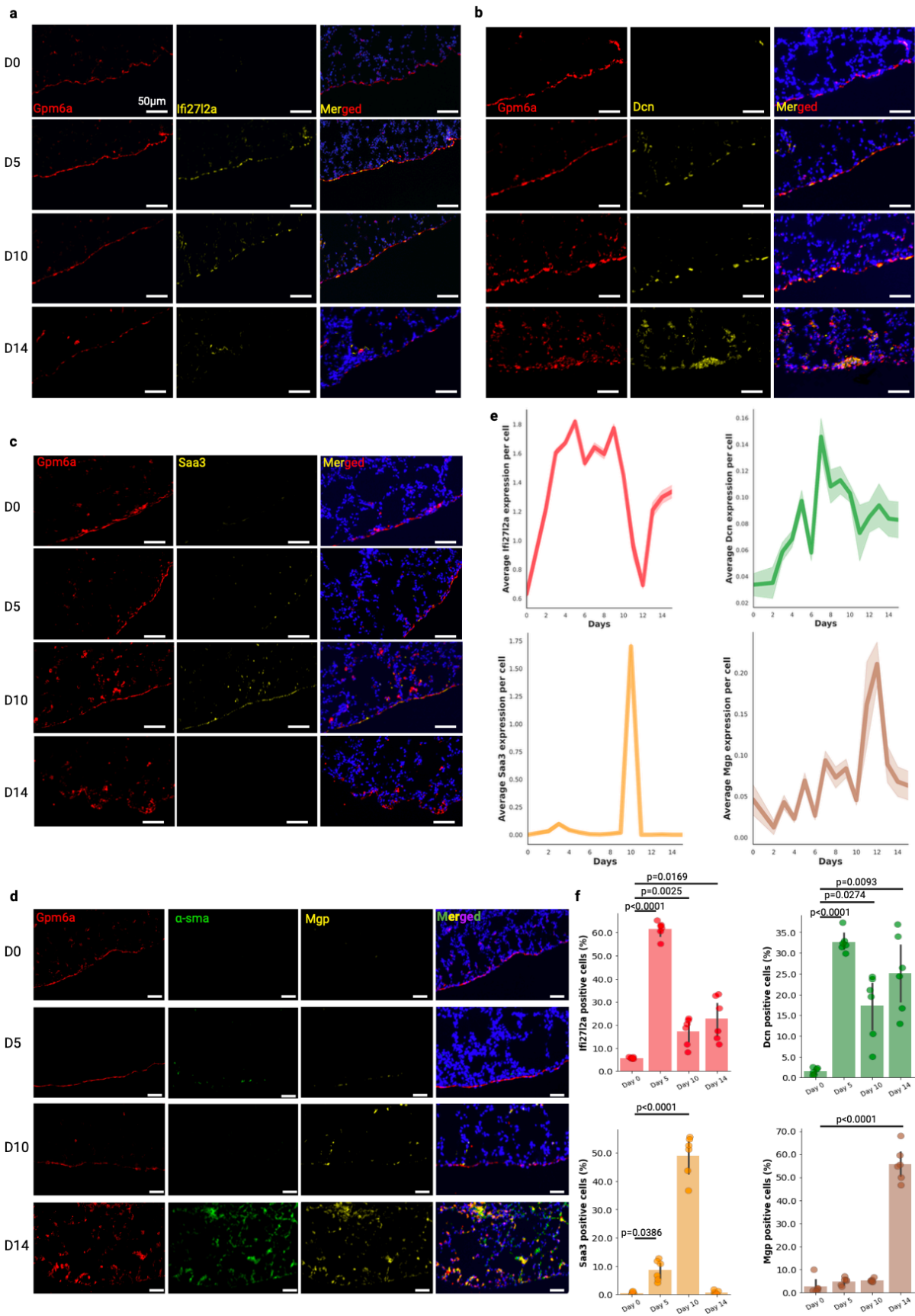
Supplementary Fig. 2



Supplementary Fig. 2: High resolution single mesothelial cell dataset in mouse lung bleomycin-induced fibrosis

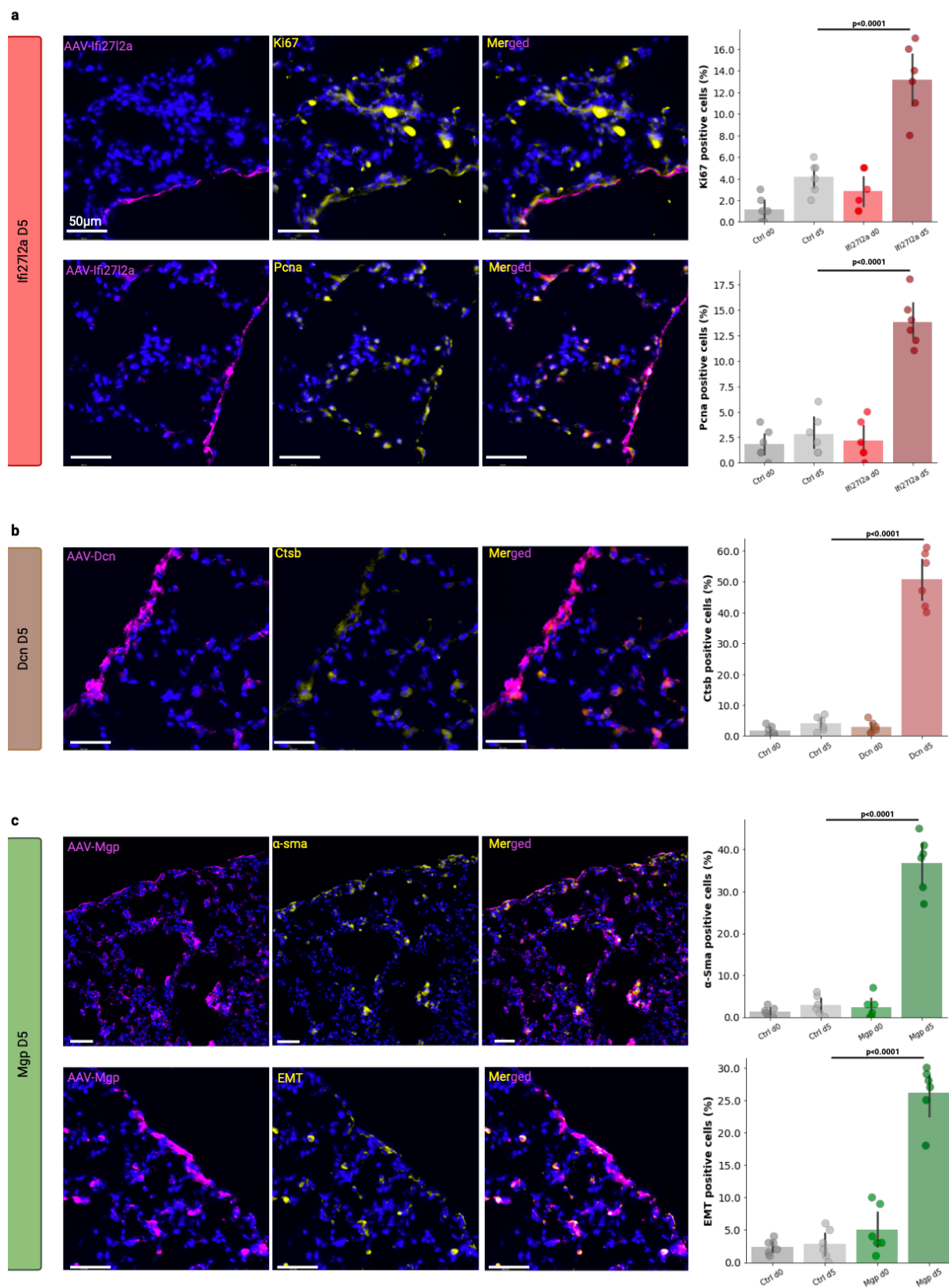
- a. UMAP embedding of mouse mesothelial single cells in lung bleomycin-induced fibrosis color coded of the expression level of enriched reactomes in the different non-healthy clusters at cellular level.
- b. UMAP embedding of mouse mesothelial single cells in lung bleomycin-induced fibrosis color coded by the newly identified clusters.
- c. Heat maps of the relative average expression of the most highly enriched genes for each cluster in the lung fibrosis mesothelial cell data ($\log(\text{fold change})$ of one cluster versus all others.
- d. UMAP embedding of mouse mesothelial single cells in lung bleomycin-induced fibrosis color coded by the different harvesting days (day0: day of bleomycin instillation)
- e. Cells fate probabilities: dots represents a cells colored by cluster labels on the left and time point at the right. Cells are arranged inside according to their fate probabilities; naive cells are placed in the middle and more fate biased cells are placed next to their corresponding corner.
- f. Top twenty-five gene markers of the five identified mouse mesothelial single cells clusters in lung bleomycin-induced fibrosis model
- g. Specific genes expression UMAP to confirm the specificity to specific clusters: Col1a1, Col1a2, Col1a3 for fibrogenic; Ctsb and MMP14 for proteolytic

Supplementary Fig. 3



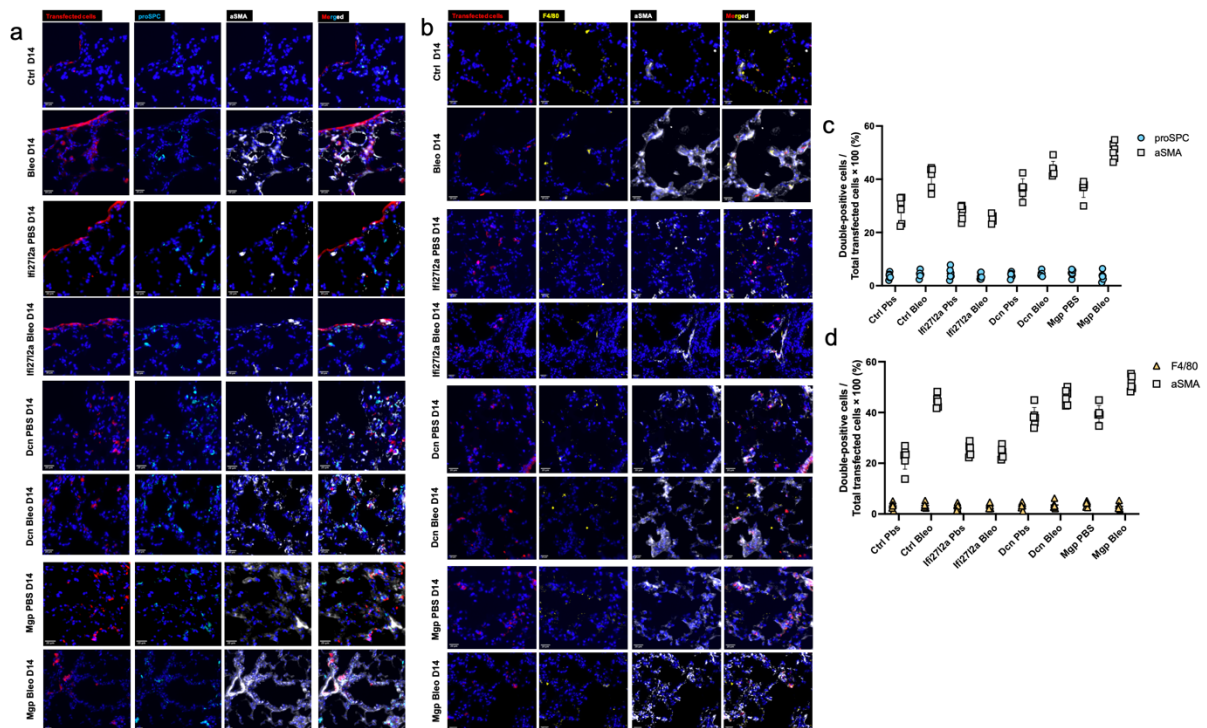
Supplementary Fig. 3: Immunostaining of the different markers identified in the high resolution lung fibrosis mesothelial cell dataset

- a. Co-Immunostaining of the metabolically active cluster marker *Ifi2712* in day 0, 5, 10 and 14 with the lung mesothelial marker *Gpm6a*. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 50µm.
- b. Co-Immunostaining of the proteolytic cluster marker *Dcn* in day 0, 5, 10 and 14 with the lung mesothelial marker *Gpm6a*. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 50µm.
- c. Co-Immunostaining of the Immune modulator cluster marker *Saa3* in day 0, 5, 10 and 14 with the lung mesothelial marker *Gpm6a*. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 50µm.
- d. Co-Immunostaining of the fibrogenic cluster marker *Mgp* in day 0, 5, 10 and 14 with the lung mesothelial marker *Gpm6a* and α -sma. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 50µm.
- e. Single cell RNA-seq average expression of the mesothelial cells from day 0 to day 14 post-bleomycin instillation of the different cluster markers: top left *Ifi2712a*, top right *Dcn*, bottom left *Saa3*, bottom right *Mgp*.
- f. Staining quantification in day 0, 5, 10 and 14 post-bleomycin instillation of the different cluster markers: top left *Ifi2712a*, top right *Dcn*, Bottom left *Saa3*, bottom right *Mgp*. A two-sided independent T-test was used for the comparison of two groups. Data are presented as mean values +/- SEM.



Supplementary Fig. 4: Immunostaining of the different markers in ex-vivo PCLS model

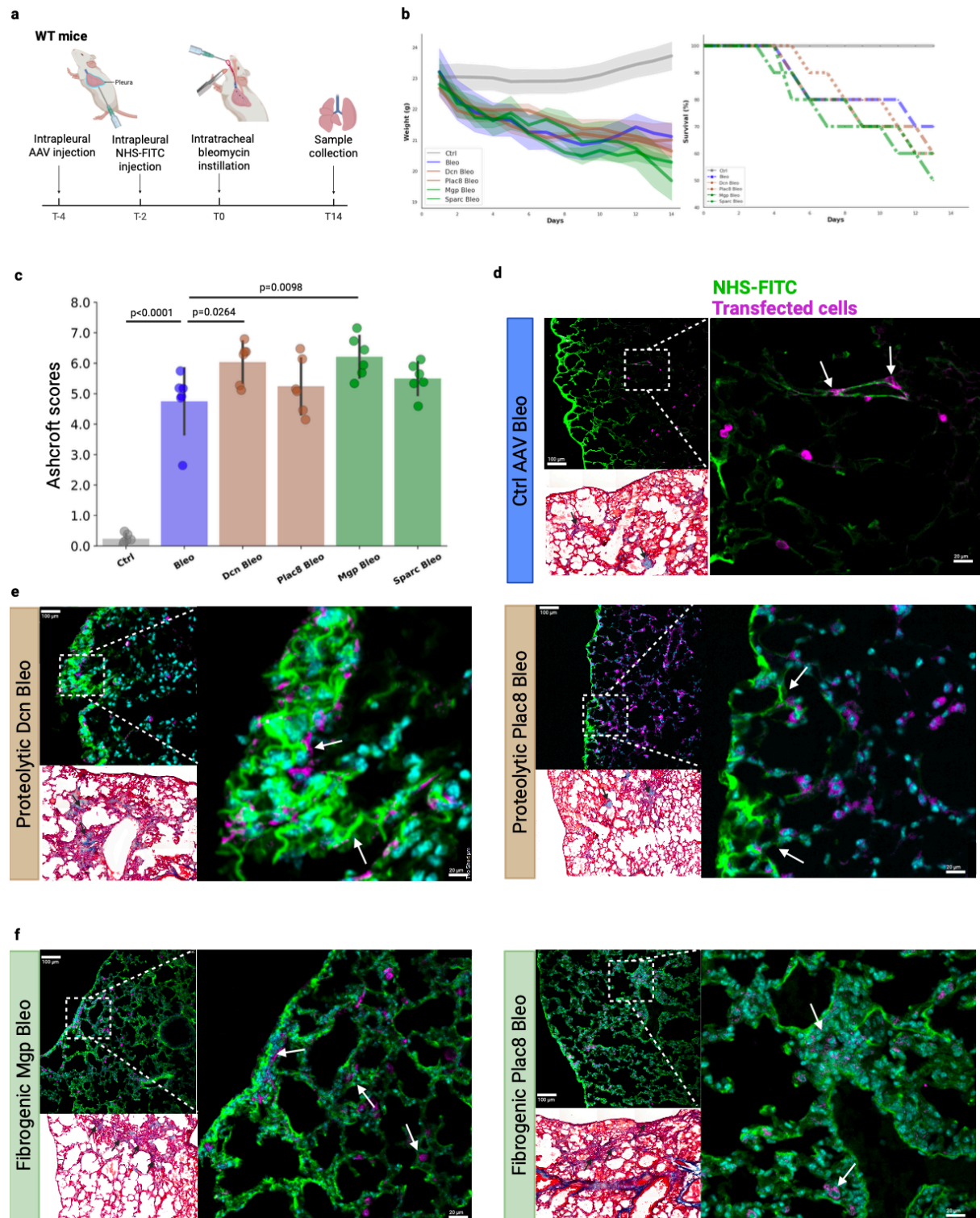
- a. Immunostaining of the mesothelial AAV-Ifi27l2a transfected PCLS for the markers Ki67 (upper panel) and PCNA (bottom panel). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. A two-sided independent T-test was used for the comparison of two groups. Data are presented as mean values $\pm$ SEM. Scale bar is 50 μ m.
- b. Immunostaining of the mesothelial AAV-Dcn transfected PCLS for the markers Ctsb. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. A two-sided independent T-test was used for the comparison of two groups. Data are presented as mean values $\pm$ SEM. Scale bar is 50 μ m.
- c. Immunostaining of the mesothelial AAV-Mgp transfected PCLS for the markers α -sma (upper panel) and EMT (bottom panel). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. A two-sided independent T-test was used for the comparison of two groups. Data are presented as mean values $\pm$ SEM. Scale bar is 50 μ m.



Supplementary Fig. 5: Immunostaining of the different markers in the mice *in-vivo* model

- Immunostaining of mesothelial cells in the AAV-transfected in-vivo model for the markers ProSPC and aSMA. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm.
- Immunostaining of mesothelial cells in the AAV-transfected in-vivo model for the markers F4/80 and aSMA. N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm.
- Quantification of the staining for the markers ProSPC and aSMA.
- Quantification of the staining for the markers F4/80 and aSMA.

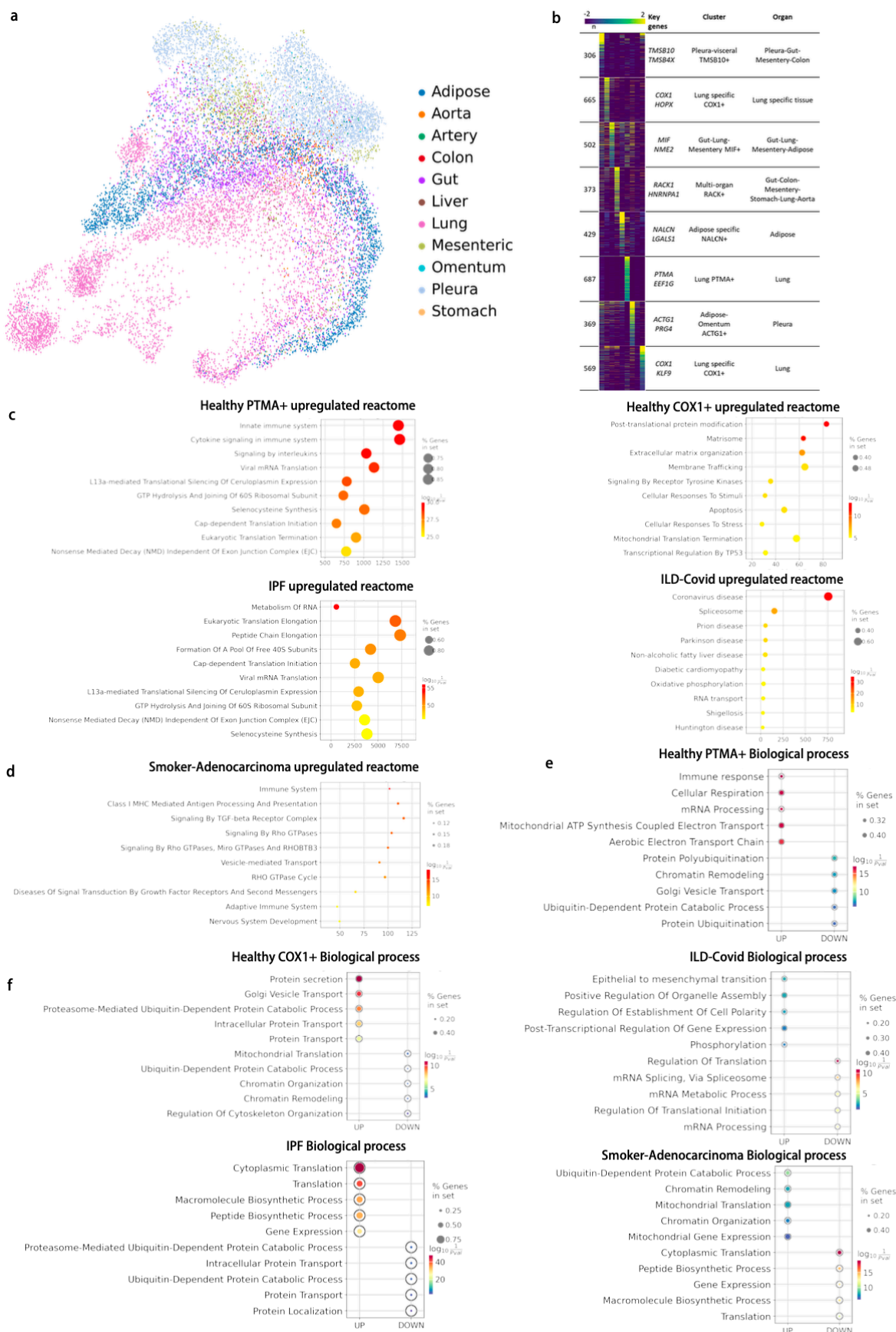
Supplementary Fig. 6



Supplementary Fig. 6: Proteolytic and fibrogenic driven mesothelial cells worsen lung fibrosis

- a. Treatment scheme for candidate gene overexpression in lung mesothelium
- b. Daily body weight lost in the different groups (left) and experimental group survival rates (right). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments.
- c. Ashcroft scores by using the trichrome staining of the lung sections in the different conditions: Control PBS (Ctrl); Control AAV Bleomycin (Bleo); Dcn AAV Bleomycin (Dcn Bleo); Plac8 AAV Bleomycin (Plac8 Bleo); Mgp AAV Bleomycin (Mgp Bleo) and Sparc AAV Bleo (Sparc Bleo). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. A two-sided independent T-test was used for the comparison of two groups. Data are presented as mean values +/- SEM.
- d. Lung function impairment in bleomycin: Trichrome staining images of the control bleomycin. Confocal images of the labeled ECM (green) and the transfected mesothelial cells (magenta). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm.
- e. Lung function impairment in proteolytic-driven mesothelial cells in bleomycin model: Trichrome staining images of the control bleomycin. Confocal images of the labeled ECM (green) and the transfected mesothelial cells (magenta). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm.
- f. Lung function impairment in fibrogenic-driven mesothelial cells in bleomycin model: Trichrome staining images of the control bleomycin. Confocal images of the labeled ECM (green) and the transfected mesothelial cells (magenta). N=6 biological replicates (C57BL/6J WT mice) and three independent experiments. Scale bar is 20µm.

Created in BioRender. Kadri, S. (2025) <https://BioRender.com/lj6t81k>



Supplementary Fig. 7: Human lung mesothelial cell atlas GO term analysis

- a. UMAP embedding of the steady-state human mesothelial cell atlas color coded by tissue of origin.
- b. Heat maps of the relative average expression of the most highly enriched genes for each cluster in steady-state human lung mesothelial atlas (log(fold change) of one cluster versus all others).
- c. Upregulated reactome of Healthy PTMA+ (top left), Healthy COX1+ (top right), IPF (bottom left) and ILD-Covid (bottom right) in the human lung mesothelial cell atlas.
- d. Upregulated reactome in Smoker-Adenocarcinoma cluster
- e. Up and down regulated biological process in Healthy PTMA+ mesothelial cell cluster
- f. Upregulated and down regulated biological process of Healthy COX1+ (top left), ILD-Covid (top right), IPF (bottom left) and Smoker-Adenocarcinoma (bottom right) in the human lung mesothelial cell atlas.

Supplementary Table 1 Disease mouse dataset

g. SRA/EMBL-EBI/DOI	Data title	GEO	tissue	Disease
E-HCAD-14	Defining the Activated Fibroblast Population in Lung Fibrosis Using Single Cell Sequencing		Lung	Fibrosis
SRA599311	The purinergic P2Y14 receptor links hepatocyte death to hepatic stellate cell activation and fibrogenesis in the liver	GSE158183	Liver	Fibrosis
SRA745744	heart healthy and heart diseased SCGs	GSE231767	Heart	transverse aortic constriction
SRA585365	Single-cell RNA-sequencing of mouse islets exposed to proinflammatory cytokines	GSE183010	Pancreas	Exposure to proinflammatory cytokines
SRA637291	IL-11 is a crucial determinant of cardiovascular fibrosis	GSE107909	Left Ventricle	myocardial infarction
SRA667466	Single-cell sequencing dissects the transcriptional identity of activated fibroblasts and identifies novel persistent distal tubular injury patterns in kidney	GSE214024	Kidney	ischemia
SRP343404	nteraperitoneal microbial contamination drives post-surgical peritoneal adhesions by mesothelial EGFR-signaling.	GSE186658	Peritoneum	Surgical adhesion
SRP234414	Alveolar regeneration through a Krt8+ transitional stem cell state that persists in human lung fibrosis	GSE141259	Lung	Fibrosis
SRP265115	ntifibrotic Drug Nintedanib Inhibits CSF1R to Promote IL-4-associated Tissue Repair Macrophage	GSE151374	Lung	Fibrosis
SRP199584	Syndecan-1 promotes lung fibrosis by regulating epithelial reprogramming through extracellular vesicles	GSE131800	Lung	Fibrosis
GSM4987107	Portal fibroblasts with mesenchymal stem cell features form a reservoir of proliferative myofibroblasts in liver fibrosis	GSE164039	Liver	Fibrosis
10.1038/s12276-021-00671-2	Single-cell transcriptome analysis reveals cellular heterogeneity in the ascending aorta of normal and high-fat diet mice		Aorta	HFD

SRP305417	Mapping the cardiac vascular niche in heart failure	GSE166403	Heart	transverse aortic constriction
SRP314466	Single-Cell Transcriptomic Analysis Reveals a Hepatic Stellate Cell-Activation Roadmap and Myofibroblast Origin During Liver Fibrosis in Mice	GSE171904	Liver	Fibrosis
SRP322180	Stiffness is associated with hepatic stellate cell heterogeneity during liver fibrosis	GSE175939	Liver	Fibrosis
E-MTAB-8483	Stromal evolution of cancer-associated fibroblasts in PDAC GEMMs		Pancreas	pancreatic ductal adenocarcinoma
SRP329005	Single-cell analysis highlights differences in druggable pathways underlying adaptive or fibrotic kidney regeneration	GSE180420	Kidney	Fibrosis
SRP280287	Single Cell RNA Sequencing Analysis of WT and CD169-DTR Adipose Tissue Vascular Fraction		vasculature	HFD
SRP292934	Loss of Fas-signaling in pro-fibrotic fibroblasts impairs homeostatic fibrosis resolution and promotes persistent pulmonary fibrosis	GSE161648	Lung	Fibrosis
SRP201481	Single cell RNA-sequencing of normal and fibrotic lungs in mouse and human	GSE132771	Lung	Fibrosis
SRP118702	Single Cell Analysis of Pulmonary Fibrotic Mesenchymal Cells Indicates a New Subtype	GSE104154	Lung	Fibrosis
SRP218543	Single-cell RNA sequencing reveals profibrotic roles of distinct epithelial and mesenchymal lineages in	GSE135893	Lung	Fibrosis

Supplementary Table 2 Healthy mouse dataset

SRA/EMBL-EBI	Data title	GEO	tissue
SRA832392	Multi-modal analysis of the aging mouse lung at cellular resolution	GSE124872	Lung
SRA599311	PDGFRA+gp38+ mesenchymal cells in the peripheral tissues support terminal differentiation of ILC2 originated from fetal liver progenitor cell	GSE102665	Mesentery
SRA628029	Neonatally imprinted mesenteric lymph node stromal cell subsets induce tolerogenic dendritic cells	GSE116633	Mesentery
E-HCAD-14	Defining the Activated Fibroblast Population in Lung Fibrosis Using Single Cell Sequencing		Lung
SCP1376	A single cell atlas of human and mouse white adipose tissue		white adipose tissue
E-MTAB-8810	Single cell RNA-Seq of the mouse heart in response to chronic stress		Heart
SRA653146	Tabula Muris: Transcriptomic characterization of 20 organs and tissues from Mus musculus at single cell resolution	GSE109774	Lung
SRA807566	Preparing for the first breath at single cell level	GSE122332	Lung
SRP284101	The purinergic P2Y14 receptor links hepatocyte death to hepatic stellate cell activation and fibrogenesis in the liver	GSE158183	Liver
SRA832392	scRNA-seq and bulk RNA-seq analysis of normoxic and hyperoxic lung in mice	GSE217488	Lung
SRA762414	Using Gjd3-CreEGFP mice to examine atrioventricular node morphology and composition	GSE118932	Heart
SRA585365	Lineage dynamics of pancreatic development at single cell resolution	GSE101099	Pancreas
SRA631205	The single cell RNA seq of pulmonary alveolar epithelial cells	GSE106960	Lung
SRA745744	Reprogram-Seq: A platform for single-cell combinatorial reprogramming	GSE117795	Whole heart
SRA753635	Single cell RNA-seq study of wild type and Hox mutant developing uterus	GSE118180	Uterus

SRA664171	Identification of Zeb1 as a single marker for prostate stem cells in the basal cell compartment	GSE111429	Prostate
E-MTAB-7149	Single cell transcriptional profiling of aortic endothelium identifies a hierarchy from endovascular progenitors to differentiated cells		Aorta
GSE180822	Three-axis classification of mouse lung mesenchymal cells reveals two populations of myofibroblasts	GSE180822	Lung
SRA837885	Adventitial stromal cells define group 2 innate lymphoid cell tissue niches	GSE125492	Adipose tissue
SRA784304	Single cell transcriptome profiling of mouse and hESC-derived pancreatic progenitors		Pancreas
E-MTAB-7703	Fibroblastic reticular cells in the spleen - total splenic Stroma cells - single cell RNAseq dataset 2	GSE120522	Spleen
SRA745744	heart healthy and heart diseased SCGs	GSE231767	Heart
SRA585365	Single-cell RNA-sequencing of mouse islets exposed to proinflammatory cytokines	GSE183010	Pancreas
SRP329005	Single-cell analysis highlights differences in druggable pathways underlying adaptive or fibrotic kidney regeneration	GSE180420	Kidney
SRP318483	Single-cell RNA-seq in AT2 cell-ablated mouse lungs	GSE173878	Lung
SRA637291	IL-11 is a crucial determinant of cardiovascular fibrosis	GSE107909	Left Ventricle
SRA665127	Single cell RNA-sequencing of visceral adipose tissue Pdgfr β + cells	GSE144299	White adipose tissue
SRA667466	Single-cell sequencing dissects the transcriptional identity of activated fibroblasts and identifies novel persistent distal tubular injury patterns in kidney	GSE214024	Kidney
SRA745567	Targeted elimination of senescent beta cells prevents Type 1 Diabetes	GSE117770	Pancreas
PRJNA975862	IL-17 signalling is critical for controlling subcutaneous adipose tissue wasting and parasite burden during chronic Trypanosoma brucei infection	GSE233312	white adipose tissue
SRP201778	A murine aging cell atlas reveals cell identity and tissue-specific trajectories of aging	GSE132901	Spleen

SRP219839	Understanding the stromal composition of fat-associated lymphoid clusters of the omentum using single-cell transcriptomics	GSE136636	Omentum
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Supplementary table 3 Human lung disease dataset

SRA/EMBL- EBI/DOI	Data title	GEO	tissue	Disease
PRJNA719842	Columbia University/NYP COVID-19 Lung Atlas	GSE171524	Lung	Covid-19+healthy
SRP395406	IPF Cell Atlas+healthy	GSE136831	Lung	IPF+Healthy
syn21041850	A molecular cell atlas of the human lung from single-cell RNA sequencing		Lung	Only healthy
SRP279837	Single cell RNA-sequencing on healthy and IPF lung mesenchymal cells.	GSE157376	Lung	IPF+Healthy
PRJNA804223	Single cell profiling of lung carcinoid tumors	GSE196303	Lung	Adenocarcinoma+Healthy
PRJNA526088	Proliferating SPP1/MERTK-expressing macrophages in idiopathic pulmonary fibrosis	GSE128033	Lung	IPF+Healthy
SRP218543	Single-cell RNA-sequencing reveals profibrotic roles of distinct epithelial and mesenchymal lineages in pulmonary fibrosis	GSE135893	Lung	ILD+Healthy
SRP261880	Transcriptional analysis of cystic fibrosis airways at single-cell resolution reveals altered epithelial cell states and composition	GSE150674	Lung	ILD+Healthy
PRJNA720539	COVID-19 tissue atlases reveal SARS-COV-2 pathology and cellular targets	GSE171668	Lung	Covid-19+healthy

PRJNA545296	Single cell RNA sequencing of lung adenocarcinoma	GSE131907	Lung	Adenocarcinoma+Healthy
SRP318548	Single-cell Transcriptome Analysis Reveals an Anomalous Epithelial Variation and Ectopic Inflammatory Response in Chronic Obstructive Pulmonary Disease	GSE173896	Lung	Smoker+Healthy
HRA002992	Single cell study of non-smoking lung adenocarcinoma		Lung	Adenocarcinoma+Healthy
10.1038/s41591-023-02327-2	An integrated cell atlas of the lung in health and disease	https://cellxgene.cziscience.com/collections/6f6d381a-7701-4781-935c-db10d30de293	Lung	ILD-IPF-Covid-Adenocarcinoma
PRJNA507000	Single-Cell Transcriptomic Analysis of Human Lung Reveals Complex Multicellular Changes During Pulmonary Fibrosis II	GSE122960	Lung	ILD-IPF-Smoker

Supplementary table 4 Human healthy

SRA/EMBL-EBI/DOI	Data title	GEO	tissue
SRP299275	Single cell RNA-sequencing of human omental adipose tissue mesothelial cells	GSE163830	Omentum
PRJNA719842	Columbia University/NYP COVID-19 Lung Atlas	GSE171524	Lung
SRP395406	IPF Cell Atlas	GSE136831	Lung
syn21041850	A molecular cell atlas of the human lung from single-cell RNA sequencing		Lung
SRP279837	Single cell RNA-sequencing on healthy and IPF lung mesenchymal cells.	GSE157376	Lung
PRJNA804223	Single cell profiling of lung carcinoid tumors	GSE196303	Lung
PRJNA526088	Proliferating SPP1/MERTK-expressing macrophages in idiopathic pulmonary fibrosis	GSE128033	Lung
SRP218543	Single-cell RNA-sequencing reveals profibrotic roles of distinct epithelial and mesenchymal lineages in pulmonary fibrosis	GSE135893	Lung
SRP261880	Transcriptional analysis of cystic fibrosis airways at single-cell resolution reveals altered epithelial cell states and composition	GSE150674	Lung
PRJNA720539	COVID-19 tissue atlases reveal SARS-COV-2 pathology and cellular targets	GSE171668	Lung
PRJNA701264	Single-cell profiling of normal and tumor intestinal tissue	GSE166555	Colon
E-MTAB-9543, E-MTAB-9536, E-MTAB-9532, E-MTAB-9533, E-MTAB-10386	Gut cell Atlas	https://www.gutcellatlas.org/	Gut

SRP336428	Cellular Atlas of Human Heart Failure	GSE183852	Heart
PRJNA636285	Understanding tumor evolution and its clonality by single-cell transcriptomic analysis in liver cancer	GSE151530	Liver
SRP340298	Single-Cell, Single-Nucleus and Spatial RNA Sequencing of the Human Liver Identifies Hepatic Stellate Cell and Cholangiocyte Heterogeneity	GSE185477	Liver
PRJNA793005	Spatial proteogenomics reveals distinct and evolutionarily-conserved hepatic macrophage niches	GSE192742	Liver
SRP281827	Human gastrointestinal epithelia of the esophagus, stomach and duodenum resolved at single-cell resolution	GSE157694	gut
https://doi.org/10.17632/y3pxvr4xbf.2	An integrated single cell and spatial transcriptomic map of human white adipose tissue	GSE225700.	white adipose tissue
PRJNA857062	Aortic Cellular Diversity and Quantitative GWAS Trait Prioritization through Single Nuclear RNA Sequencing (snRNA-Seq) of the Aneurysmal Human Aorta	GSE207784	Aorta
PRJNA871957	Single-cell RNAseq of mesenteric adipose tissue from Crohn's disease patients and healthy controls	GSE215001	Mesentery
SRP197094	Single cell transcriptome of peritoneal cells	GSE130888	Pritoneum
SRP281827	Human gastrointestinal epithelia of the esophagus, stomach and duodenum resolved at single-cell resolution	GSE157694	Stomach