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Reporting Summary

Nature Portfolio 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 Portfolio policies, see our Editorial Policies 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	No code was used for data collection.
Data analysis	Single-cell RNA sequencing reads were processed with Cell Ranger on the Terra platform (https://app.terra.bio/). The count matrices obtained from Cell Ranger were uploaded into R (v4.2.3) to remove the ambient RNA contamination using the SoupX package v1.6.2. The resulting matrices were used as input to construct a Seurat object using the Seurat package v4. The identification of putative multiplets was done using the scDblFinder v1.12.0 and the DoubletFinder v2.0.4 packages. Cells labeled as multiplets by any of the two methods were removed from the count matrices. The Seurat v4 package was used for further filtering low-quality cells. The resulted Seurat objects were integrated using CCA integration in Seurat v4. We employed CellChat (v2.1.2) to infer cell-cell interactions. For STARmap, images for individual tiles were deconvoluted using Huygens (23.04). Image registration was performed using 3D Fourier Transform through numpy.fft and Scipy. Starfish v0.2.2 was used to process sequencing signals. StarDist (0.9.1) was employed for 2D cell segmentation of identified transcripts. Segmented cells from each sample were integrated for subsequent single-cell analysis using Seurat v4. Label transfer was performed using Seurat v4. Imputation was performed using Tangram (1.0.4). Additional data processing and visualization were performed using NumPy (v2.2.6), Pandas (v2.3.3), SciPy (v1.15.3), scikit-image (v0.25.2), Matplotlib (v3.10.8), and Scanpy (v1.11.5). Code for comprehensive Raman and Raman analysis as described here, and figure generation as shown here can be found at https://github.com/jian-shu-lab/RamanOmics

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 and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio 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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

All data generated in this study are deposited in the SenNet Consortium data portal (https://data.sennetconsortium.org/) under the data group "TDA-Massachusetts Institute of Technology" and are publicly available at: https://doi.org/10.60586/SNT495.DLVB.649
Dataset identifiers are as follows: snRNA-seq: SNT283.FCFK.385 (mouse lung), SNT497.WWSP.546 (mouse skin); STARmap: SNT928.MXCF.423 (mouse lung), SNT963.RGBR.439 (mouse skin), SNT937.VCKK.546 (mouse skin, wound healing); Raman hyperspectral imaging: SNT459.BDNV.556 (mouse lung), SNT467.XLCG.675 (mouse skin), SNT598.LRRF.663 (mouse skin, wound healing).

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	N/A
Reporting on race, ethnicity, or other socially relevant groupings	N/A
Population characteristics	N/A
Recruitment	N/A
Ethics oversight	N/A

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

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	We collected naturally aged mouse skin and lung tissues from young (2-month-old, n= 3) and aged (26-month-old, n= 3) mice, as well as wound-healing skin samples from aged (24-month-old, n= 3) mice. (in total, n= 9 mice). No statistical methods were used to pre-determine sample sizes; n=3 biological replicates per group is consistent with sample sizes used in comparable single-cell and spatial transcriptomic profiling studies of mouse lung and skin (Joost et al., Cell Stem Cell, 2020; Angelidis et al., Nat. Commun., 2019)
Data exclusions	Single cells during single-cell analysis were excluded based on stringent filtering criteria (e.g., high mitochondrial reads, high/low gene/UMI counts).
Replication	All key findings (differential gene expression, differential Raman peaks, multimodal classifier performance) were consistent across the n=3 biological replicates per group. No experiments failed to replicate.
Randomization	Samples were allocated to experimental groups based on predefined biological variables, including age, tissue type, and injury status. No randomization was performed, as group assignment was inherent to the study design rather than treatment-based.
Blinding	Blinding researchers would not have any productive effect on this study; thus, blinding was not utilized and is not relevant to this study

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 and archaeology
☐	☒ Animals and other organisms
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

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

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals	p16-3MR mice (Mus musculus, congenic on a C57BL/6 background) were obtained from Jackson Lab (RRID: IMSR_JAX:037045) and housed under a 12h light/12h dark cycle at 22±2°C with 40-60% relative humidity and ad libitum access to food and water. For single-cell transcriptomics, spatial transcriptomics, and Raman imaging experiments, young mice were 2 months old (n=3, all male) and aged mice were 26 months old (n=3, two male and one female). For wound-healing experiments, mice were 24 months old (n=3, all male)
Wild animals	This study did not involve wild animals
Reporting on sex	Both male and female mice were included in the aging cohort (2 male, 1 female for old mice; all male for young mice); all wound-healing cohort mice were male. Sex was recorded at the time of sample collection but was not used as a stratification factor in the experimental design, and no sex-specific analyses were performed due to limited sample size
Field-collected samples	This study did not involve samples collected from the field.
Ethics oversight	All animal experiments were approved by the Institutional Animal Care and Use Committee (IACUC#2018N000182) at Massachusetts General Hospital, in accordance with the relevant animal husbandry standards of the Committee on Animal Care

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

Plants

Seed stocks	N/A
Novel plant genotypes	N/A
Authentication	N/A