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Corresponding author(s): Kuei-Pin Chung, Ching-Hua Kuo, Chong-Jen Yu

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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	n/a
Data analysis	1. FlowJo analytical software (version 10; BD Biosciences) was used for analyzing all flow cytometric data. 2. Densitometry of immunoblots was performd using FIJI running Image J software (version 1.51w). 3. All results of extracellular flux analyses were analyzed using the Wave software (version 2.6.3.5; Agilent Technologies) 4. The alveolar mechanic properties (lung function) were retrieved from flexiWare software (SCIREQ). 5. For analyzing RNAseq data, the cutadapt (v3.5)(https://github.com/marcelm/cutadapt) was applied to remove adaptor sequences and to filter out the low-quality reads. The resulting clean reads were then aligned to the mouse genome assembly (GRCm38.p6) by using STAR (v 2.7.8a)(https://github.com/alexdobin/STAR) with two-pass mode. To quantify gene expression, a gene count matrix was generated based on the GENCODE vM25 gene annotation using featureCounts (v2.0.1)(http://subread.sourceforge.net). To account for differences in library size between the samples, the trimmed mean of M values (TMM) normalization method implemented by edegR (https://bioconductor.org/) was applied, and the abundance of transcripts was measured with transcripts per million (TPM). For identifying differentially expressed genes, the procedures implemented by limma package (https://bioconductor.org/) was employed. The p values were computed using linear modelling and empirical Bayes moderation, and were adjusted for multiple comparison through the Benjamini-Hochberg procedure. Over-representation analyses of the differentially expressed genes were performed using the R package clusterProfiler (https://bioconductor.org/). The analyses focused on Gene Ontology (GO) Biological Process sets were obtained from MSigDB (v7.0, C5). The significantly enriched GO sets (adjusted $p < 0.05$) were utilized to construct association networks using enrichmentMap (https://apps.cytoscape.org/apps/enrichmentmap), and networks were visualized by Cytoscape (version 3.7.0) (https://cytoscape.org/). Heat maps were plotted using the pheatmap package (https://cran.r-project.org/web/packages/pheatmap/), based on the z scores calculated using the gene expressions by log2TPM. The upstream regulator analysis was performed using Ingenuity Pathway Analysis (IPA) (Qiagen). 6. For scRNAseq datasets downloaded from Gene Expression Omnibus, data pre-processing, including data integration, feature selection,

normalization, and data reduction was conducted using the Scanpy (v1.9.0).
7. For stable isotope resolved metabolomics, data from UHPLC-QTOF mass spectrometry were analyzed using Agilent MassHunter Workstation Profinder Software (Version B.10.1) with an in-house database established by Agilent Personal Compound Database and Library. The results exported by Profinder were corrected for the natural distribution of 13C.
8. For lipidomic measurements, data from UHPLC-QTOF mass spectrometry were analyzed by using MassHunter Workstation Profinder Software Version B.08.00 (Agilent Technologies). MassHunter Workstation Quantitative Analysis Software Version B.07.01 (Agilent Technologies, Santa Clara, CA, USA) was used for peak integration of the data from UHPLC-triple quadrupole mass spectrometry.
9. For metabolomic analyses using BALF samples, all the peaks were integrated with MassHunter Quantitative Analysis software.
10. All statistical analyses were performed using GraphPad Prism version 10.0.0 (GraphPad Software, San Diego, CA, USA) or SPSS version 18.0.0 (IBM, Armonk, NY, USA).

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

RNA sequencing data are available under the accession number GSE224354 and GSE255949 in Gene Expression Omnibus (GEO). Source data underlying Fig. 1b-c, e-f, 2a, c-h, 3a-k, 4b-c, e, g, 5b-h, 6b, d, e, h-l, 7g-k, 8e-m, and Supplementary Fig. 4a-d, f, g, 6b, 7b-i, 8b-c, 9, 10a, 11b, 12b, 13, 15b-m, 19, 20c, 23a-d, 24a, and 4c are provided with this paper as the Source Data file. All the microscopy images in the figures and supplementary figures, including Fig. 1a, 7a, and Supplementary Fig. 1, 2, 10b, 21 and 22a, have been deposited in Mendeley online data repository (https://doi.org/10.17632/343cym3z8p.1). Requests for resources reagents and materials should be directed to the lead contact, Kuei-Pin Chung (gbchung@ntu.edu.tw). All the materials and MS-associated raw data generated in this study are available from the lead contact on reasonable request.

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	Demographic data, including age and biological sex, of each enrolled subject were collected. Data about biological sex in two clinical cohorts are summarized and reported in Supplementary Table. Sex-based analyses are not performed in this study, since ARDS develops in both male and female patients.
Reporting on race, ethnicity, or other socially relevant groupings	Not applicable
Population characteristics	1. For the immunohistochemistry study cohort, the study population is constituted by adult subjects (age >= 20 years) with diffuse alveolar damage and surgical lung samples from January 2004 through November 2020, and those who received lung segmentectomy or lobectomy for lung adenocarcinoma in National Taiwan University Hospital from September through November in 2021. 2. For BALF cytokine profiles and PG contents, the study population is constituted by adult subjects with ARDS who were admitted to medical intensive care units at National Taiwan University Hospital and Fu Jen Catholic Hospital from May 2020 through December 2022, and those with GOLD grade 0-1 chronic obstructive pulmonary disease at the out-patient clinics of National Taiwan University Hospital Yunlin branch.
Recruitment	1. For the immunohistochemistry staining study cohort, the participants were retrospectively identified from January 2004 through November 2020 according to the database of the Pathology Index System, Department of Pathology, National Taiwan University Hospital. Waiver of informed consent was approved by the Institutional Review Board at National Taiwan University Hospital (protocol 202012115RIND) 2. For BALF cytokine profiles and PG contents, participants were recruited in medical intensive care units at National Taiwan University Hospital and Fu Jen Catholic Hospital, from May 2020 through December 2022. The participants in the control group were recruited at the outpatient clinics of National Taiwan University Hospital Yunlin Branch, from Aug 2019 through Feb 2022. All the subjects were recruited and enrolled after informed consents were obtained (protocol 201911058RINA, 202712075RINA and FJUH109018).
Ethics oversight	The protocols for all the human studies above were approved by the Institutional Review Board at National Taiwan University Hospital and National Taiwan University Hospital Yunlin branch (202012115RIND, 201911058RINA, and 201712075RINA), and at Fu Jen Catholic Hospital (FJUH109018).

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	Sample sizes for all experiments were made as large as possible to ensure the robustness of the results, under the consideration of 3R (reduction, refinement, replacement) policy. All the details about the sample size for each experiment (n number information) were described in the figure legends.
Data exclusions	No data were excluded.
Replication	All the reported findings are based on multiple biological and/or technical repeats, and the numbers of replicates are described in the figure legends. All attempts at replication were successful for all the data reported in this study.
Randomization	The grouping is based on the genotypes of the mice, and randomization is not applicable.
Blinding	The experimental conditions of the animals were known to the investigators. The blinding to the experimental conditions is difficult since ALI will lead to gross changes of murine lungs.

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

Antibodies

Antibodies used	1. IHC study: Primary antibodies against human CPT1a (1:10; catalog 12252, Cell Signaling Technology), LCAD (1:50; catalog ab152160, Abcam) and CK (PCK26, pre-diluted, Roche VENTANA ULTRA kit, Roche Diagnostics) 2. Isolation of murine AT2 cells: anti-CD45 microbeads (catalog 130-052-301, Miltenyi Biotec), and biotin-conjugated anti-EpCAM antibody (catalog 13-5791-82, eBioscience) 3. Immunoblots: CPT1a (1:1000; catalog ab128568, Abcam), LCAD (1:1000; catalog ab129711, Abcam), SFTPC (1:1000; catalog ABC99, Merk Millipore), TOM20 (1:1000; catalog sc11415, Santa Cruz,Dallas, TX, USA), DDK/FLAG (1:1000; catalog TA50011-100, OriGene), and β-actin (1:5000; catalog MAB1501, Merk Millipore) 4. Flow cytometric analyses of immune cells in BALF: anti-CD45.2-FITC (1:100; catalog 553772, BD Biosciences), anti-CD11c-PE-Cy7 (1:80; catalog 25-0114-81, eBioscience), anti-Ly6G-Alexa Fluor 700 (1:200; catalog 127621, BioLegend), anti-CD3-PE-Cy5 (1:200; catalog 555276, BD Biosciences), and anti-SiglecFPE (1:100; catalog 552126, BD Biosciences) 5. Mitochondrial reactive oxygen species in AT2 cells: anti-EpCAM-PE-Cy7 (1:150; catalog 25-5791-80, eBioscience) 6. Immunofluorescent staining using cryosections of murine lungs: primary antibodies against CXCL1 (1:100; catalog MAB4532-SP, R&D systems), and CXCL2 (1:100; catalog AF-452-SP, R&D systems), and CD45 (conjugated to Alexa Fluor 647, 1:100; catalog 103123, BioLegend); secondary antibodies against goat IgG (conjugated to Alexa Fluor 488; 1:500; catalog A11055, Thermo Fisher Scientific) and rabbit IgG (conjugated to Alexa Fluor 488; 1:500; catalog A-11008, Thermo Fisher Scientific) 7. Immunofluorescent staining using human surgical lung samples: primary antibodies against CPT1a (1:10; catalog 12252, Cell Signaling Technology), HTII-280 (1:50; TB-27AHT2-280, Terrace Biotech); secondary antibodies against rabbit IgG (conjugated to Alexa Fluor 647; 1:500; A31573, Thermo Fisher Scientific) and mouse IgM (conjugated to Alexa Fluor 488; 1:500; catalog A-21042, Thermo Fisher Scientific).
Validation	1. The antibodies for AT2 isolation and flow cytometric analyses: Nat Commun 2019; 10: 3390 (DOI: 10.1038/s41467-019-11327-1) 2. Flow cytometric analyses of immune cells in BALF: J Vis Exp 2017; 123: e55498 (DOI: 10.3791/55398).

3. Antibodies used for immunoblots, IHC staining, and immunofluorescent staining were validated in the immunoblots (anti-CPT1a, Abcam ab128568, in Fig. 2c and Supplementary Fig. 7a; anti-LCAD, Abcam ab129711, in Fig. 8e) in this study, or in manufacturers' websites.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	MLE12 and HEK293T cell lines were all obtained from ATCC.
Authentication	No specific authentication was performed.
Mycoplasma contamination	The cell lines have been tested and are free from mycoplasma infection.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

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	All the mouse lines in this study were maintained in National Laboratory Animal Center (NLAC), NARLabs (Taipei, Taiwan) and in Laboratory Animal Center, National Taiwan University College of Medicine (Taipei, Taiwan). Both animal centers are accredited by the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) International. All the experimental mice were bred in the individually ventilated cages with autoclaved bedding, and ad libitum fed with sterilized water and irradiated PicoLab® Rodent Diet 20 (catalog 5053, LabDiet, St Louis, MO, USA) in Laboratory Animal Center, National Taiwan University College of Medicine. Cpt1aloxP/loxP (Cpt1atm1.1Pec) mice were generated and provided by Peter Carmeliet, Vlaams Instituut voor Biotechnologie. SftpcCreERT2+/+ (B6.129S-Sftpctm1(cre/ERT2)Blh/J) mice and ROSA26tdTomato+/+ (B6.Cg-Gt(ROSA)26Sortm14(CAG-tdTomato)/Hze/J) mice were provided by Augustine MK Choi, Weill Cornell Medicine. C57BL/6JNarl mice were obtained from National Laboratory Animal Center (NLAC), NARLabs (Taipei, Taiwan). AcadlloxP/loxP (B6-Acadlem2CJY/J) mice were generated by the CRISPR (clustered regularly interspaced short palindromic repeats)/Cas9 technology by the Transgenic Mouse Model Core Facility of the National Core Facility for Biopharmaceuticals, National Science and Technology Council, Taiwan, and the Animal Resources Laboratory of National Taiwan University Centers of Genomic and Precision Medicine.
Wild animals	No wild animals were used in the experiments.
Reporting on sex	Both male and female mice were used for animal experiments. Mice matched for ages and sex, both male an female, were used for alveolar metric analyses (10~12 weeks), histology examination (~37 weeks) and models of ALI (10~12 weeks). Data from both male and female mice were reported together.
Field-collected samples	No field-collected samples were used in this study.
Ethics oversight	The animal experiments and procedures were approved by the Institutional Animal Care and Use Committee (IACUC) at National Taiwan University College of Medicine and College of Public Health (protocols 20180128, 20201154, 20210138, 20210401, and 20230151).

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

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration	Not applicable. This study did not involve clinical trial.
Study protocol	Not applicable. This study did not involve clinical trial.
Data collection	Only demographic data, including age and biological sex, of each enrolled subject were obtained.
Outcomes	Not applicable

Plants

Seed stocks	Not applicable
Novel plant genotypes	Not applicable
Authentication	Not applicable

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	Please refer to Methods for the details about preparing whole lung single cell suspension, isolating murine AT2 cells, collecting bronchoalveolar lavage fluid samples, and labeling newly generated choline-containing phospholipids through CLICK labeling.
Instrument	Flow cytometric analysis and sorting were performed using a BD LSRII flow cytometer or a BD FACSAria III flow cytometer.
Software	The results were analyzed using FlowJo analytical software (version 10)
Cell population abundance	The cell population abundance depends on the samples for cell cytometric analyses. Specific cell populations were identified through surface marker staining and flow cytometric identification.
Gating strategy	FSC and SSC were applied to set the gate for single cell population. For flow cytometric experiments using live cells, DAPI staining was applied to exclude dead cells. The gating strategies and algorithm were reported together with the flow cytometric results in Figures and Supplementary Figures.

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