

Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure 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 |
|-------------------------------------|--|
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The statistical test(s) used AND whether they are one- or two-sided <i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> A description of all covariates tested |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> 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) |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> For null hypothesis testing, the test statistic (e.g. F , t , r) with confidence intervals, effect sizes, degrees of freedom and P value noted <i>Give P values as exact values whenever suitable.</i> |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> 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

Data analysis

All 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](#)

The authors declare that the data supporting the findings of this study are available within the paper and its Supplementary Information Files. Source data are provided as source data files. The source data of statistical analysis in the figures are provided as a Source Data file 1. The source data of uncropped scans of Western-blot in the figures are provided as a Source Data file 2. Mass-spectrum analysis refer to Uniprot database and the source data are provided as a source data file 3.

Human research participants

Policy information about [studies involving human research participants and Sex and Gender in Research](#).

| | |
|-----------------------------|----|
| Reporting on sex and gender | NA |
| Population characteristics | NA |
| Recruitment | NA |
| Ethics oversight | NA |

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](https://www.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 | The cell samples were collected and performed with at least three independent experiments. The samples from animal were collected from three mice every group and performed with three independent experiments. The human samples were independently collected for at least three times and measured with two duplicates. The exact size was described in the figure or the legend. For animal experiments, we choose same age of control and experiment littermate mice. We measure the weight of body, spleen and other organs. For the cellular experiments, we count the cell number and use the same amount of control and experiment cells to do further experiments. |
| Data exclusions | No data were excluded in the analysis in this study. |
| Replication | All experimental findings described in this study were repeated at least three times and got the consistent results before make relative conclusions. |
| Randomization | The samples/organisms/participants were randomly allocated into experimental group. |
| Blinding | All the authors involved in the experiments in this study were blinded to group allocation during data collection and/or analysis. |

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 | Involvement in the study |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Antibodies |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Eukaryotic cell lines |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Palaeontology and archaeology |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Animals and other organisms |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Clinical data |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Dual use research of concern |

Methods

| | |
|-------------------------------------|--|
| n/a | Involvement in the study |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> ChIP-seq |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Flow cytometry |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> MRI-based neuroimaging |

Antibodies

| | |
|-----------------|---|
| Antibodies used | The following antibodies were used in western blotting experiments: Antibodies against RIPK1 (3493), phospho-RIPK1 (31122), cleaved Caspase3 (9664), Caspase3 (9665), cleaved Caspase8 (9429), Caspase8 (4790), A20 (5630), p-IkB α (2859), p-p38 (9211), p38 (9228), p-JNK (9251), JNK (9252), p-TBK1 (5483), TBK1 (3504), JAK1 (50996), p-JAK1 (74129), STAT1 (14994), p-STAT1 (9167), SRC |
|-----------------|---|

(2109), p-MK2 (3007), MK2 (3042), cFLIP (56343) and RelB (4954) were purchased from Cell Signaling Technology; Antibodies against RIPK3 (sc-374639), I κ B α (sc-1643), PCNA (sc-56), FADD (sc-6036) and p65 (sc-109) were purchased from Santa Cruz Biotechnology; Antibodies against phospho-MLKL (ab196436) and MLKL (ab67942); Antibodies against β -tubulin (BE0025-100), GFP (BE2002), GAPDH (BE0023), secondary horseradish peroxidase (HRP)-conjugated anti-rabbit (BE0108-100) and anti-mouse antibodies (BE0107-100) were obtained from Easy Bio; Antibody against phosphor-tyrosine (4G10) were purchased from Millipore; Antibody against Flag (M20008), HA (M20006) were purchased from Abmart. AF700 anti B220 (48-0452-80), V450 anti-CD3 (48-0032-8246-0041-82), Percp-cy5.5 anti-CD4 (46-0041-82), FITC anti-CD8 (11-0081-82), FITC anti-CD11b (11-0112-82), PE anti-F4/80 (12-4801-82), PE anti-c-Kit (12-1171-81), Percp-cy5.5 anti-CD16/32 (45-0161-82) and V500-conjugated Cell viability Dye(65-0866-18) were purchased from eBioscience; Percp-cy5.5 anti-Ly6C (561237), V450 anti-Ly6G (560603), FITC anti-CD34 (553733) antibodies were from BD Bioscience; APC anti-CD71 (113819), PE anti-Ter119 (116207), PB anti-Sca-1 (108120), APC anti-CD150 (115909) and FITC anti-Lineage (133313) antibodies were from BioLegend. Antibodies for western-blotting were used with 500-fold or 1000-fold dilution and for flow cytometry were used with 200-fold dilution.

Validation

All antibodies used in this study were validated according to the manufacturer's instruction and all worked well.

Eukaryotic cell lines

Policy information about [cell lines and Sex and Gender in Research](#)

Cell line source(s)

The source of HEK293T cells were from ATCC, and the source of immortalized MEFs and BMDMs were described in the methods.

Authentication

The HEK293T cells were purchased from ATCC and authenticated by morphology and the vendor, and the immortalized MEFs were authenticated according to the genotypes of embryos.

Mycoplasma contamination

We declare that all cell lines used in this research are mycoplasma negative by experimental detection.

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

Research animals used in this study are RIPK1 Y383F, Tnfr1^{-/-}, Ripk3^{-/-} and Caspase-8^{-/-} mouse lines on a C57BL6/J background. These mice are housed in the specific pathogen-free (SPF) animal facilities (light/dark cycle 10h:14h, temperature 22-26°C, humidity 40%-70%).

Wild animals

The study did not involve wild animals.

Reporting on sex

The study did not involve sex-based analysis.

Field-collected samples

The study did not involve samples collected from the field.

Ethics oversight

All mouse experiments were performed in compliance with institutional guidelines and according to the protocol approved by the Institutional Animal Care and Use Committee of Tsinghua University.

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

For mice samples, the spleens from mice were perfused and digested into single-cell suspensions. After RBC lysis buffer treatment, the whole cells were washed with PBS and then stained with corresponding fluorescent antibodies and left at 4°C until use. All mouse experiments were performed in compliance with institutional guidelines and according to the protocol approved by the Institutional Animal Care and Use Committee of Tsinghua University.

Instrument

For cell analysis, LSRFortessa (BD Biosciences) was used.

| | |
|---------------------------|---|
| Software | Flow cytometry data were analyzed with FlowJo (Tree Star). |
| Cell population abundance | Without using sorting strategy. |
| Gating strategy | In the spleen or bone marrow of mice, the living cell fractions gated from preliminary FSC/SSC gates could be further divided into four cell types: CD4+ T cells, CD8+ T cells, CD19+B220+ B cells, CD11b+Ly6G+ neutrophils, CD11b+F4/80+ macrophage, CD11b+Ly6C+ monocyte, Ter119+ red blood cells, Ter119intCD71int erythroblast R1, Ter119intCD71high erythroblast R2, Ter119highCD71high erythroblast R3, Ter119highCD71int erythroblast R4, Ter119highCD71low erythroblast R5, Lin-Sca-1+Kit+ (LSKs) progenitors, Lin-Sca-1-Kit+ (LKs) progenitors, LSK-CD150+CD34- Hematopoietic stem cells, Lin-Sca-1-Kit+FcyR+CD34+(GMPs) progenitors, Lin-Sca-1-Kit+FcyR-CD34+ (CMPs) progenitors and Lin-Sca-1-Kit+FcyR-CD34- (MEPs) progenitors. All this is shown in supplementary Figures and information. |

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