nature portfolio

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Last updated by author(s): Jan 21, 2025

# 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<br>*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<br>*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 | Image Lab 3.0 (Bio-Rad), CytExpert 2.3.0 (Beckman coulter). |
| Data analysis | FlowJo 10.8 (BD Bioscience), ImageJ 1.46r (NIH), PyMOL 2.6 (DeLano Scientific LLC), GraphPad Prism 9.0 (GraphPad Software Inc) and SPSS 20.0 software (IBM). |

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 analyzed during this study are included in this paper and its supplementary files. The datasets analyzed for this study can be found in the Gene Expression Omnibus (GEO) of NCBI (https://www.ncbi.nlm.nih.gov/geo/) (Accession: GSE280007). Supporting data related to this work are available upon 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 | Both male(n=6) and female(n=6). Gender was determined based on self-reporting. |
| Reporting on race, ethnicity, or other socially relevant groupings | Han ethnic group is the main people of china, and the participants were Han people. |
| Population characteristics | See Supplementary Table 1. Patient demographics. |
| Recruitment | Random selection among surgical patients following patient consent. |
| Ethics oversight | Renmin Hospital of Wuhan University (approval number: WDRY2021-KS047) |

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 | The sample size was based on previous studies. |
| Data exclusions | No data were excluded from analysis. |
| Replication | All experiments were performed independently at least three times with similar results, as described in the figure legends. |
| Randomization | Random number method used to group mice. Proteins and cells were randomly allocated to the wells in each experimental group. |
| Blinding | For histology analysis, technicians performing the experiments and collecting the data were given the ID of the mice but not the group. Random allocation and quantitative measurements using various approaches minimized biased assessments. |

# 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 | EZH2 CST #5246<br>α-SMA CST #19245<br>Collagen I Affinity AF7001 |

Fibronectin CST #26836
GAPDH GAPDH GB15004-100
DUSP23 Genetex 108245
H3K27me3 ABclonal A2363
H3 ABclonal A17562
SMAD3 CST #9523
pSMAD3-Ser423/425 CST #9520
pSMAD3-T179 Abcam 74062

Validation

All antibodies used in this study are commercially available and have been validated by the manufacturers' and/or previous publications.

## Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

| | |
|---|---|
| Cell line source(s) | Mice macrophage cell line (RAW264.7, male) was purchased from Procell Life Science&Technology Co.,Ltd. |
| Authentication | Cell lines were verified by manufacturer's website or published papers and Identity of these cell lines were frequently checked by their morphological features. |
| Mycoplasma contamination | All cell lines were tested to be mycoplasma-negative by PCR. |
| Commonly misidentified lines (See ICLAC register) | No commonly misidentified cell lines are used in this study. |

## 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 | EZH2fl/fl mice (Cre-negative, C57BL/6J background) were obtained from Pro. Xi Wang (Department of Immunology, School of Basic Medical Sciences; Advanced Innovation Center for Human Brain Protection, Beijing Key Laboratory for Cancer Invasion and Metastasis, Department of Oncology, Capital Medical University, Beijing, China.) and CAG-creER mice were acquired from our previous studies42–44. CAG-creEREZH2fl/fl (EZH2.iKO) mice were conducted at six weeks of age with 75 mg/kg tamoxifen intraperitoneal injection (T5642; Sigma-Aldrich) for five days.The mice were acclimatized with free access to water and normal chow. Male mice were selected for model construction in order to improve the success rate of the model.<br>Male C57BL/6 mice (weight 24-26 g, 6-8 weeks old, n=6, each group) were obtained from Animal Experiment Center at the Renmin hospital of Wuhan University. |
| Wild animals | No wild animals were involved. |
| Reporting on sex | Male mice were selected for model construction in order to improve the success rate of the model. |
| Field-collected samples | No field-collected samples were involved. |
| Ethics oversight | Laboratory Animal Welfare & Ethics Committee of the Renmin Hospital of Wuhan University (approval number: WDRM-20200604). |

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 involved. This is an observational study. |
| Study protocol | We collected kidney specimens from patients with nephrocalcinosis-related CKD (n=6, Supplementary Table.1). Non-tumoral adjacent kidney tissues were used as a control group (n=6). We used immunofluorescence (IF) to identify MMT in kidney tissues by the coexpression of myofibroblast markers (collagen I or α-SMA) and macrophage markers (CD68). |
| Data collection | Kidney specimens were taken immediately after the nephrectomy. |
| Outcomes | This is an observational study. |

## Plants

| | |
|---|---|
| Seed stocks | NA |
| Novel plant genotypes | NA |
| Authentication | NA |

## 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 | Macrophages were isolated from kidneys in mice by ficoll-gradient centrifugation and fluorescence-activated cell sorting (FACS). Briefly, kidney tissues were minced and sieved with 70μm cell filter. Cellular suspension was added to a 10ml tube with ficoll (1.084 g/ml) and centrifuged at 400×g for 30 min. In vitro, cells were incubated with antibodies and applied to Flow Cytometry. |
| Instrument | BeckmanCytoFLEX. |
| Software | Flowjo 10.8 software, CytExpert 2.3.0. |
| Cell population abundance | Total of 15,000-20,000 cells |
| Gating strategy | Presented in Fig. 6B. Cells were identified by APC/PE/BV421. |

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