nature portfolio

Corresponding author(s): Laurie Ailles

Last updated by author(s): Aug 12, 2024

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	Native software on Illumina Novaseq6000 S4 system and native software on Illumina Novaseq X were used for RNAseq data collection; Incucyte base analysis software was used for cell proliferation data collection; BD FACS Diva software was used for flow cytometry data collection; Zen software on a Zeiss AxioImagerI fluorescence microscope was used for fluorescence microscopy image collection; CFX Manager software (Bio-Rad CFX96 real-time PCR detection system) was used for qRT-PCR data collection. Native software on Q-Exactive HF (Thermo Scientific) mass spectrometer was used for Bio-ID data collection; Cell quantification in initial epiprobe screen was done using LICOR acquisition software.
Data analysis	RNAseq data analysis: TrimGalore v0.6.6 (cutadapt v3.0 and fastqcv0.11.5), STAR v2.7.9a, HTSeqv0.11.0, DESeq R package v1.36.0, PANTHER17.0, rMATS v4.3.0 Bio-ID mass spectrometry data analysis: Proteowizard (v3.0.19311), X!Tandem (v2013.06.15.1), Comet (v2014.02.rev2), transproteomic pipeline (PP v4.7), Significance Analysis of INTeractome (SAINT), Cytoscape 3.0.1, STRING Enrichment app. Fluorescence images: Image J (foci detection); Image J with the OpenComet plugin v1.3 for Comet assay. Flow cytometry analysis: FlowJo v. 10.7.1 Q-RT-PCR data analysis: Microsoft Excel Statistics: GraphPad Prism v9.3.1

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

The data supporting the findings of this study are available within the paper and its supplementary information files. Source data for figures and supplementary figures are provided as a Source Data file. The raw and processed RNAseq data generated in this study have been deposited in NCBI's Gene Expression Omnibus and are accessible through GEO Series accession number GSE229357. The proximity labeling proteomics data generated in this study have been deposited in ProteomeXchange under accession code MSV000094655.

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	No sex/gender based analyses were performed
Reporting on race, ethnicity, or other socially relevant groupings	N/A
Population characteristics	N/A
Recruitment	ccRCC samples used to generate patient-derived cell lines were obtained from the University Health Network and the Cooperative Health Tissue Network (CHTN) from patients providing written consent.
Ethics oversight	University Health Network Research Ethics Board

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	Statistical methods were not used to predetermine sample size. For the analysis of fluorescence microscopy images (foci and comet assay tail moments), cell numbers ranging from 16 to 147 cells per condition were analyzed, providing sufficient power to identify differences between conditions. For in vitro and in vivo studies, exact numbers are reported in figure legends and were sufficient to identify statistically significant differences.
Data exclusions	No data were excluded
Replication	Experiments were repeated at least two times or as stated in figure legends and all attempts at replication were successful.
Randomization	For in vivo experiments, mice were randomized into treatment groups. For other experiments, samples were randomly allocated into treatment groups (e.g. replicates of cell lines treated with drug vs DMSO followed by various assays).
Blinding	Investigators were not blinded during data collection and analysis as experimental setup and analyses were performed by the same individuals.

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

PRMT1: Thermo, PA5-17299, 1:1000
Asymmetric dimethylarginine: Sigma, #09-814, 1:1000
Tubulin: Santa Cruz #sc-5286, 1:1000
GAPDH: Santa Cruz, #sc-69778, 1:1000
Histone H4R3me2a: Active Motif, #39705, 1:1000
Histone H4: Abcam, #ab174628, 1:1000
BUB1B: Cell Signalling Technology, #4116S, 1:1000
CENPI: Cell Signaling Technology, #49426S, 1:1000
CENPA: Cell Signaling Technology, #2186T, 1:1000
Actin: Abcam #ab7817, 1:1000
phospho-histone H3: Millipore, #06-570, 1:1000
BRCA2: Cell Signaling Technology, #107415, 1:1000
RAD51AP1: Thermo/Proteintech, #11255-1-AP, 1:1000
FANCM: Bethyl Labs, #A302-637A, 1:1000
FANCD2: Cell Signaling Technology, #16323, 1:1000 (for Westerns) or 1:100 (for immunofluorescence)
H2A.X: Millipore, #05-636, 1:1000
S9.6: Fischer Scientific, #MABE1095MI, 1:100
BCLAF1: Thermo/Bethyl Labs, #A300-608A, 1:1000
CAS9: Abcam, #ab191468, 1:1000
FLAG: Sigma, F3165, 1:1000
Streptavidin-HRP: BD Pharmingen, #554066, 1:1000
anti-rabbit IgG-HRP: Cell Signaling Technology, #7074S, 1:2500
anti-mouse IgG-HRP: Cell Signaling Technology, #7076S, 1:1000
anti-Rabbit IgG H+L -Alexa FluorTM 594: Invitrogen #A11037, 1:400
anti-Mouse IgG (H+L) -Alexa FluorTM 488: Invitrogen #A10667, 1:1000

Validation

All antibodies were purchased from commercial vendors who provide validation information on their website, including published references. In addition, the following antibodies were validated using genetic tools in our laboratory: PRMT1, H4R3me2a and asymmetric dimethyl-arginine (aDMA).

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)

786-0 cells were obtained from the American Type Culture Collection (ATCC CRL-1932).
Remaining cell lines were derived from patient samples were derived from patient samples in our own laboratory as listed below:
RCC22 (female, age 68)
RCC162 (male, age 59)
RCC222 (male, age 49)
RCC243 (female, age 48)
RCC323 (male, age 57)
RCC364 (male, age 45)
RCC407 (male, age 62)
Methods for cell line derivation are described in Lobo et al, BMC Cancer. 2016 Jul 16;16:485. doi: 10.1186/s12885-016-2539-z

Authentication

All cell lines were authenticated by STR profiling.

Mycoplasma contamination

All cell lines were confirmed to be negative for mycoplasma

Commonly misidentified lines
(See ICLAC register)

N/A

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	For xenograft studies male or female NOD.Cg-Prkdc<scid>Il2rg<tm1WjI>/SzJ (NSG) mice between 6 and 12 weeks old were used as recipients. Mice were bred in-house and housed in ventilated cages maintained at 50% humidity (+5%/-10% deviation) with ambient temperature of 72oF (+/- 2oF deviation) and a 12 hour ligh/dark cycle.
Wild animals	N/A
Reporting on sex	N/A
Field-collected samples	N/A
Ethics oversight	All animal experiments were approved by the Institutional Animal Care and Use Committee (IACUC) at the University Health Network.

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

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	At the time of collection cells were trypsinized with 0.25% trypsin and washed with media and PBS. Cells were then fixed in paraformaldehyde and resuspenced in FACS buffer (Hanks balanced salt solution with with 2% FBS).
Instrument	Becton Dickonson LSR2 analytical flow cytometer
Software	BD FACS Diva software was used to collect data and FlowJo v. 10.7.1 was used for data analysis.
Cell population abundance	Only analytical flow cytometry was done, no cell sorting was done. A minimum of 10,000 events was collected. GFP-positive populations ranged from 0 to 100%
Gating strategy	Cells were first gated on forward scatter (FSC) and side scatter (SCC) to exclude debris. Cells were then gated by FSC-height vs FSC-width and SSC-heigh vs SSC-width to esclude doublets. Next, viable cells were excluded on a FSC vs Zombie fixable violet dye (Biolegend, 423114), and two-parameter density plots (FSC vs GFP) were then used to identify GFP+ vs GFP- cells.

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