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Reporting Summary

Nature Research 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 Research policies, see Authors & Referees 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	All software is commercially or freely available. Focinator V 2.0 was used to analyze Immunfluorescence images. Statistical Analysis was done in PRISM 7 or Stata. Figures were compiled using Adobe Illustrator. For LC/MS assays, Analyst® 1.6 software was used for system control and data acquisition and MultiQuant® 3.0 software was used for data processing and quantitation. Flow cytometry data was collected using FlowJo CE and analyzed using FlowJo™ v7.6.5 software.
Data analysis	All software is commercially or freely available. Focinator V 2.0 was used to analyze Immunfluorescence images. Statistical Analysis was done in PRISM 7 or Stata. Figures were compiled using Adobe Illustrator. Analyst® 1.6 software was used for system control and data acquisition and MultiQuant® 3.0 software was used for data processing and quantitation. Flow cytometry data was analyzed using FlowJo™ v7.6.5 software. For ChIP-seq analysis, raw sequencing data was aligned to the GRhg38 (hg38) genome build using Bowtie2 (v2.3.4.1). H3K9me3 peaks were called using HOMER (v4.10) using findPeaks with options -style histone -size 1000 -minDist 2500, filtered by P = 0.01.

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/reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research 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 list of figures that have associated raw data
- A description of any restrictions on data availability

All data generated or analyzed during this study are included in this published article (or its supplementary information files, including source data). There are no restrictions on data availability.

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	For in vitro assays, n=3 biological replicates were used for sample size in all cases order to allow statistical analyses. Each biological replicate was defined as an independent culture of cells. No statistical methods were used to determine sample size. For the in vivo mouse studies, mice were randomized into irradiated and mock irradiated groups with 8 mice/group, based on a calculated power of 80% to detect a difference of 1.25 standard deviations between two groups at 5% significance level.
Data exclusions	No data were excluded.
Replication	Replicates are indicated in the manuscript with n=3 biological replicates for experimental data presented here. All replicates were successful.
Randomization	Cultures and mice were randomly assigned to either experimental or control groups.
Blinding	In vitro assays were not conducted in a blinded manner. Xenograft tumors were randomly assigned into treatment or control groups. Tumor growth measurements were not conducted in a blinded fashion. Blinding was not relevant to our study, since subjective rating of data was not involved.

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
☐	☒ Animals and other organisms
☒	☐ Human research participants
☒	☐ Clinical data

Methods

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

Antibodies

Antibodies used

anti-ATM (phospho S1981) (abcam, ab36810)
anti-ATM(Millipore, Ab-3)
anti ß-Actin HRP (Proteintech, HRP-60008)
anti-Histone 3 Lysine 9 trimethyl (D4W1U, Cell Signaling Technology)
anti-Histone H3 (# 9715 Cell Signaling)
anti-Histone H3 (ab1791, Abcam)
Rabbit anti-HA-Tag (C29F4, Cell Signaling Technology)
mouse anti- GAPDH HRP (HRP-60004, Proteintech),
mouse anti-IDH1 R132H (H09, Dianova)
anti-IDH1 (D2H1, Cell Signaling Technology)
anti-Fumarase (D9C5, Cell Signaling Technologies)
anti-SDHB (21A11AE7, Abcam)
anti-RAD51(14B4 Novus bio)
anti-BRCA2 (ab1, Millipore)
ati-TI60 (NBP2-24613. Novus)
anti-RPA32 (#52448, Cell Signaling Technology)
anti-MRE11 (H-300, Sant Cruz Biotechnology sc-22767)
anti-Phospho-(Ser/Thr) ATM/ATR Substrate Antibody (Cell Signaling Technology, #2851)
anti-KDM4A (A300-861A ,Bethyl)
anti-KDM4A (C70G6, Cell Signaling Technology)
anti KDM4B (A301-478A, Bethyl) 1:1000 5% BSA

anti-KDM4B (D7E6, Cell Signaling Technology)
anti-Ku80 80 (Clone 7, BD Biosciences)
anti-Poly-ADP-Ribose (PAR) (4335-mc-100, Trevigen)
anti-phospho-RPA32 (E5A2F, Cell Signaling Technology)
Mouse anti-CHK1 (2G1D5, Cell Signaling Technology)
anti-phospho-CHK1 (#2341 Cell Signaling Technology)
anti-phospho-CHK2 (C13C1, Cell Signaling Technologies)
anti-HIF1a (NB100-105, Novus)
anti-Vinculin (ab129002, abcam)
anti Histone H2A.X (D17A3, Cell Signaling Technology)
anti DNA-PKcs (ab70250, abcam)
anti phosphor-DNA-PKcs (ab 18192, abcam)
anti-H3K36me3 (D5A7, Cell Signaling Technologies)
anti-H3K27me3 (C36B11, Cell Signaling Technologies)
Rabbit anti-H3K4me3 (C42D8, Cell Signaling Technologies)

Antibody dilutions for western blots were as follows: mouse anti-ATM (phospho S1981) antibody (Abcam, ab36810) 1:100 in 5% milk, rabbit anti-ATM (Millipore, Ab-3) 1:1000 in 5% milk, mouse anti-ß-Actin HRP (Proteintech, HRP-60008) 1:1000 in 5% milk, anti-Histone 3 Lysine 9 trimethyl (D4W1U, Cell Signaling Technology) 1:1000 in 5% BSA, rabbit polyclonal anti-Histone H3 (ab1791, Abcam) 1:5000 in 5% BSA, rabbit anti-HA-Tag (C29F4, Cell Signaling Technology) 1:2000 in 5% BSA, mouse anti-GAPDH HRP (HRP-60004, Proteintech), mouse anti-IDH1 R132H (H09, Dianova) 1:2000 in 5% milk, rabbit anti-IDH1 (D2H1, Cell Signaling Technology) 1:1000 in 5% BSA, rabbit monoclonal anti-Fumarase (D9C5, Cell Signaling Technologies) 1:1000 in 5% BSA, mouse monoclonal anti-SDHB (21A11AE7, Abcam) 1:1000 in 5% milk, mouse monoclonal anti-RAD51(14B4 Novus bio) 1:1000 in 5% milk, mouse anti-BRCA2 (ab1, Millipore) 1:1000 in 5% milk, rabbit anti-T60 (NBP2-24613, Novus Biologicals) 1:1000 in 5% BSA, rabbit anti-RPA32 (#52448, Cell Signaling Technology) 1:1000 in 5% BSA, Rabbit anti -MRE11 (H-300, Sant Cruz Biotechnology sc-22767) 1:1000 in 5% milk, rabbit anti- Phospho-(Ser/Thr) ATM/ATR Substrate Antibody (#2851,Cell Signaling Technology) 1:1000 in 5% BSA, rabbit anti-KDM4A (A300-861A, Bethyl) 1:1000 in 5% BSA, rabbit anti-KDM4A (C37E5, Cell Signaling Technology) 1:1000 in 5% BSA, anti-KDM4B (A301-478A, Bethyl) 1:1000 in 5% BSA, anti-KDM4B (D7E6, Cell Signaling Technology) 1:1000 in 5% BSA, rabbit anti-FLAG (SAB4301135, Sigma) 1:1000 in 5% BSA, rabbit anti-SUV39H1 (D11B6, Cell Signaling Technology) 1:1000 in 5% BSA, mouse anti-Ku80 80 (Clone 7, BD Biosciences) 1:1000 in 5% Milk, rabbit anti-Histone H2A.X (D17A3, Cell Signaling Technology) 1:1000 in 5% BSA, mouse anti-Poly-ADP-Ribose (PAR) (4335-mc-100, Trevigen), mouse anti-phospho-ATM S1981 (ab36810, Abcam) 1:1000 in 5% Milk, rabbit anti-phospho-RPA32 (E5A2F, Cell Signaling Technology) 1:1000 in 5% BSA, mouse anti-CHK1 (2G1D5, Cell Signaling Technology) 1:1000 in 5% BSA, rabbit anti-phospho-CHK1 (#2341, Cell Signaling Technology) 1:1000 in 5% BSA, rabbit anti-CHK2 (#2662, Cell Signaling Technologies) 1:1000 in 5% BSA, rabbit anti-phospho-CHK2 (C13C1, Cell Signaling Technologies) 1:1000 in 5% BSA, rabbit anti-DNA-PKcs (ab70250, Abcam) 1:1000 in 5% milk, rabbit anti phospho-DNA-PKcs (ab 18192, Abcam) 1:1000 in 5% milk, mouse anti-HIF1a (NB100-105, Novus) 1:1000 in 5% BSA, rabbit anti-H3K36me3 (D5A7, Cell Signaling Technologies) 1:1000 in 5% BSA, rabbit anti-H3K27me3 (C36B11, Cell Signaling Technologies) 1:1000 in 5% BSA, rabbit anti-H3K4me3 (C42D8, Cell Signaling Technologies) 1:1000 in 5% BSA.

Validation

Antibody validation appears in Extended Data Figure 3. We used siRNA to knockdown the target proteins and validated knockdown by western blot, then we performed the DSB-ChIP protocol in the cells with the target protein suppressed to validated the ChIP signal from these antibodies. Knockdown of the H3K9 specific methyltransferase SUV39H1 served to validate the H3K9me3 antibody. All antibodies used in this study are commercially available, and were otherwise validated by the manufacturer, by previous studies from other laboratories or by previous studies from our laboratory, as cited in the text and methods.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)

YUNK1 cells were derived by us and have been previously described (6). They were generated in culture from uninvolved cortical renal tissue from a patient undergoing a radical nephrectomy for kidney cancer. They were immortalized with a lentiviral vector containing SV40 Large T antigen (Addgene plasmid #22298). YUNK1 cells with shRNA suppression of SDHB or FH using the GIPZ lentiviral constructs (GE Dharmacon) have been previously described (6). shRNA suppression of SDHB and FH in the HEK293FT cells (obtained from ATCC) were achieved using TRIPZ lentiviral knockdowns and these cells are previously described (6). Hela cells were obtained from ATCC. Heterozygous IDH1 R132H/+ HeLa cells were derived by us and have been previously described (5). HCT116 cells and HCT116 IDH1 R132H/+ cells were obtained from Horizon Discovery (Waterbeach, UK). Immortalized astrocytes expressing WT or mutant IDH1 have been previously described (9,28) and were a gift from T. Chan (Sloan Kettering). U87 IDH1 WT and IDH1 R132H/WT glioblastoma cells were obtained from the ATCC. SN1079 and RBE cells were obtained from the RIKEN cell bank. SW1353 cells were obtained from ATCC. UOK 262 cells were a gift from M. Linehan and have been previously described (29). NCCFH1 were obtained from B.T. Teh. and have been previously described (30).

Authentication

Cell lines were authenticated using STR analysis.

Mycoplasma contamination

Cell were routinely tested for mycoplasma and tested negative.

Commonly misidentified lines
(See ICLAC register)

No commonly misidentified cell lines were used in this study.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	Female athymic nu/nu mice (Hsd:Athymic Nude-Foxn1nu, Envigo) were used at 8 weeks of age.
Wild animals	not applicable
Field-collected samples	not applicable
Ethics oversight	The authors confirm that all animal experiments were performed in accordance with relevant guidelines and regulations. All studies were approved by the Yale University Institutional Animal Care and Use Committee.

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

ChIP-seq

Data deposition

☒ Confirm that both raw and final processed data have been deposited in a public database such as GEO.

☒ Confirm that you have deposited or provided access to graph files (e.g. BED files) for the called peaks.

Data access links *May remain private before publication.*	We did not generate ChIP-seq data in this study. Rather, we analyzed published and publicly available data. Raw ChIP-seq data corresponding to dox-induced IDH1 R132H mutant (MUT) and uninduced parental (PAR) astrocyte cell lines after 40 passages in culture, as well as corresponding input data, were accessed from publicly available SRA data (SRP082568) from experiments previously published in Turcan, S. et al. Mutant-IDH1-dependent chromatin state reprogramming, reversibility, and persistence. Nat Genet 50, 62-72, doi:10.1038/s41588-017-0001-z (2018).
Files in database submission	Not applicable. See comment above.
Genome browser session (e.g. UCSC)	not applicable

Methodology

Replicates	Not applicable. See comment above.
Sequencing depth	Not applicable. See comment above.
Antibodies	Not applicable. See comment above.
Peak calling parameters	Not applicable. See comment above.
Data quality	Not applicable. See comment above.
Software	See comment above. For ChIP-seq analysis, data from Turcan, S. et al. Mutant-IDH1-dependent chromatin state reprogramming, reversibility, and persistence. Nat Genet 50, 62-72, doi:10.1038/s41588-017-0001-z (2018) was aligned to the GRhg38 (hg38) genome build using Bowtie2 (v2.3.4.1). H3K9me3 peaks were called using HOMER (v4.10) using findPeaks with options -style histone -size 1000 -minDist 2500, filtered by P = 0.01.

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 the EJ2 and EJ5 assays, reporter cells were washed with PBS, collected by trypsinization, then assayed for GFP expression by flow cytometry. For cell cycle analysis, cells were fixed with cold ethanol for 1 h at 4C, then stained with Propidium iodide solution.
Instrument	Becton Dickinson FACSCalibur

Software	Data was collected with FlowJo CE and data was analyzed with FlowJo version 7.6.5 software.
Cell population abundance	Cell populations for EJ2 and EJ5 were differentiable by GFP expression above 10^3 intensity and quantified with FlowJo.
Gating strategy	For GFP analysis in the EJ2 and EJ5 assay, the GFP threshold for gating was established using the positive control samples and set at 10^3 intensity. This gating strategy is shown in the supplementary Information. A negative control sample confirmed this gating strategy, before the exact same gate was applied to all experimental samples.

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