

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 (<i>n</i>) 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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☐ | ☒ 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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted
<i>Give P values as exact values whenever suitable.</i> |
| ☒ | ☐ 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 <i>d</i> , Pearson's <i>r</i>), 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	Live imaging software: CQ1 software version 1.05.01.02
Data analysis	Statistical analyses were performed with GraphPad Prism software (version 9.0) Live imaging software: CQ1 software version 1.05.01.02 ImageJ version 1.51f used for image analysis Statistical analysis was performed using R v4.1.3 Chromothripsis was called using CTLPScanner available at: http://cailab.labshare.cn/CTLPScanner/download.php Alignment was performed with the EnsembleVariantCallingPipeline: https://github.com/AlexandrovLab/EnsembleVariantCallingPipeline Alignment itself was performed with BWA-mem v0.7.17 BAM files were sorted using samtools v1.9 Duplicates were identified with Picard v2.18.15 MarkDuplicates EnsembleVariantCallingPipeline v1.0 Copy number calling was performed using ASCAT v3.1.0 (https://github.com/VanLoo-lab/ascats) Structural variant calling was performed using delly v1.1.6 (https://github.com/dellytools/delly) Structural variant VCFs were converted to bedpe files using bcftools v1.17-5-g36f1b0c R libraries: GenomicRanges v1.46.1 beanplot v1.3.1 qvalue v2.26.0 ggplot2 v3.3.6 scales v1.2.0

ASCAT v3.1.0
ComplexHeatmap v2.12.1
circlize v0.4.15

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](#)

Whole genome sequencing data (FASTA files) from RPE1 cell lines generated for this study are available through SRA with accession number PRJNA961206.

The human GRCh38.d1.vd1 was used as a reference sequence, downloaded from: <https://gdc.cancer.gov/about-data/gdc-data-processing/gdc-reference-files>

TCGA gene expression data is available from:
<https://portal.gdc.cancer.gov>

PCAWG chromothripsis calls are available from:
https://dcc.icgc.org/releases/PCAWG/evolution_and_heterogeneity/clustered_mut_processes

Processed copy number profiles from SNP6 arrays for TCGA are available from:
https://github.com/VanLoo-lab/ascats/tree/master/ReleasedData/TCGA_SNP6_hg19

Human research participants

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

Reporting on sex and gender

Population characteristics

Recruitment

Ethics oversight

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

Data exclusions

Replication

Randomization

Blinding

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

Methods

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

Antibodies

Antibodies used

Anti-phospho histone H2AX (Ser139) (gammaH2AX) (clone JBW301, EMD Millipore, 05-636, 1:1500 IF) (Cell Signaling Technologies, 2577, 1:1000 IF), anti-TOPBP1 (Bethyl Laboratories, A300-111A, 1:2000 IF, 1:2000 IB) (Abcam, ab2402, 1:1000 IF), anti-MDC1 (Abcam ab11171, 1:1000 IF, 1:1000 IB), anti-CIP2A (clone 2G10-3B5; Santa Cruz sc80659, 1:500 IF, 1:1000 IB), anti-GAPDH (clone 14C10, Cell Signaling Technologies, 1:5000 IB), anti-PolD3 (clone 3E2, Abnova, H00010714-M01, 1:1000 IB), anti-Lig4 (clone N2C2, GeneTex, GTX100100, 1:1000 IB), anti-Mre11 (clone 12D7, GeneTex, GTX70212, 1:1000), anti-Rad50 (Cell Signaling Technologies, 3427, 1:1000 IB), anti-HA-tag (Novus biologicals, NB600-363, 1:2000 IB), anti-Lamin A/C (clone E-1, Santa Cruz, sc-376248, 1:200 IF), anti-Lamin B1 (clone C-5, Santa Cruz, sc-365962, 1:200 IF), anti-53BP1 (Thermo Fisher Scientific, PA1-16565, 1:1000 IF), anti-BRCA1 (clone D-9, Santa Cruz, sc-6954, 1:1000 IF). Secondary antibodies: Rb Alexa Fluor 488 (Thermo Fisher, A-11008, 1:1000 IF), Ms Alexa Fluor 488 (Thermo Fisher, A-11001, 1:1000 IF), Rb Alexa Fluor 568 (Thermo Fisher, A-11011, 1:1000 IF), Ms Alexa Fluor 568 (Thermo Fisher, A-11004, 1:1000 IF), Rb Alexa Fluor 647 (Thermo Fisher, A-31573, 1:1000 IF), Ms Alexa Fluor 647 (Thermo Fisher, A-31571, 1:1000 IF).

Validation

All antibodies used are commercially available and have been used by numerous papers. Validation data is available on the manufacturers website.

Eukaryotic cell lines

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

Cell line source(s)

DLD1 cells (Cleveland Lab stock, ATCC).
293T cells (Cleveland Lab stock, ATCC).
RPE1 WT cells (Cleveland Lab stock, ATCC).
RPE1 p53^{-/-} cells (Gift from Prof. David Pellman).
RPE1 MDC1^{-/-} cells (Gift from Prof. Manuel Stucki).
RPE1 CIP2A^{-/-} cells (Gift from Prof. Manuel Stucki).
RPE1 H2AX S139A/S139A (Gift from Prof. Steve Jackson).

Authentication

DLD1 cells were authenticated previously by sequencing and cytogenetic analysis.
293T cells authenticated by their culture behavior (loosely attached), morphology under the microscope, and by their efficiency in transfection and virus production.
RPE1 p53^{-/-} cells, RPE1 MDC1^{-/-} cells, RPE1 CIP2A^{-/-} cells, RPE1 H2AX S139A/S139A cells were authenticated by their morphology, culture behavior, no other authentication procedure was used.

Mycoplasma contamination

All cell lines used in this study were confirmed to be free of mycoplasma contamination, by routine (every 6 months) testing and by frequent DAPI staining and Plasmotest Reagent Kit (InvivoGen).

Commonly misidentified lines (See [ICLAC](#) register)

No commonly misidentified cell lines were used in this study.