

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	N/A
Data analysis	We have used the latest version available of the following algorithms: BEDtools v2.29.1, https://github.com/XiaoTaoWang/TADLib Calder v0.7, https://github.com/CSOgroup/CALDER2 Chimera-X v1.3, https://www.cgl.ucsf.edu/chimerax/ Chrom3D, https://github.com/Chrom3D/pipeline Cogent NGS analysis pipeline v2.0.1, https://www.takarabio.com/learning-centers/next-generation-sequencing/bioinformatics-resources/cogent-ngs-analysis-pipeline coolpup.py v0.9.5, https://coolpuppy.readthedocs.io/en/latest deepTools v3.5.1, https://deeptools.readthedocs.io/en/develop/ DESeq2 v1.36.0, https://bioconductor.org/packages/release/bioc/html/DESeq2.html eCLIP analysis pipeline, https://github.com/YeoLab/eCLIP FIJI colocalization plugin, https://imagej.nih.gov/ij/plugins/rgb-profiler.html HiTAD v0.4.5-r1, https://github.com/open2c/cooltools HTSeq v0.12.4, https://github.com/simon-anders/htseq IsoformSwitchAnalyzeR v2.2.0, https://www.bioconductor.org/packages/release/bioc/html/IsoformSwitchAnalyzeR.html Juicebox, https://www.aidenlab.org/juicebox/ Mayavi, https://pypi.org/project/mayavi/ Metascape, https://metascape.org/ Napari, https://github.com/napari/

NGseqBasic pipeline, <https://github.com/Hughes-Genome-Group/NGseqBasic>
 Picard tools v2.20.7, <https://github.com/broadinstitute/picard>
 R programming v4.1.0, <https://cran.r-project.org/bin/windows/base/old/>
 RUVseq v1.30.0, <https://github.com/drisso/RUVSeq>
 Salmon v1.5.0, <https://salmon.readthedocs.io/en/latest/salmon.html>
 scikit-image, <https://scikit-image.org/>
 STAR v2.7.3a, <https://github.com/alexdobin/STAR>
 Whippet v1.6, <https://github.com/timbitz/Whippet.jl>

All original code used for Chrom3D modeling is available at: <https://github.com/varamojo/SimChrom/>; that for Molecular Dynamics simulations is available at: <https://github.com/marianoimperatore/BPDL>; and that for LoopTrace data analysis is available at: <https://git.embl.de/grp-ellenberg/looptrace>.

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](#)

Data availability

All NGS data generated in this study have been deposited to the NCBI Gene Expression Omnibus (GEO) and are available under the following accession numbers GSE269856 [RNA-seq], GSE269857 [eCLIP], GSE269858 [ChIPmentation], and GSE283687 [APEX2-seq]. This paper also analyzes publicly available GEO data: GSE98448 [HUVEC RNA-seq; ref.8], GSE113957 [skin fibroblast RNA-seq; ref.73], GSE171782 [HMGB2 sCLIP; ref.14], GSE154095 [IMR90 TSA-seq; ref.35], GSE238256 [Micro-C and CTCF CUT&Tag; ref.38], and human whole blood RNA-seq was shared upon request (from ref.64).

Code availability

All original code used for Chrom3D modeling is available at: <https://github.com/varamojo/SimChrom/>

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

We have demonstrated similar effects in both female (IMR90) and male human primary cells (HUVECs). Other than that, gender-specific differences were not taken into account in studying this ubiquitous senescence-linked phenotype.

Reporting on race, ethnicity, or other socially relevant groupings

N/A

Population characteristics

N/A

Recruitment

N/A

Ethics oversight

N/A

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

No statistical method was used to predetermine sample size, but sample sizes and replicates are in line with what is typically used in the field (e.g., experiments performed in independent triplicates).

Data exclusions

No data were excluded from analyses.

Replication	All experiments were performed in at least three independent replicates, unless otherwise stated. Attempts at replication were successful.
Randomization	N/A; experiments were not randomized.
Blinding	The Investigators were not blinded to allocation during experiments and outcome assessment, with the exception of RNA-seq data that were pseudonymized prior to data 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	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

Primary:

anti-BANF1, Abcam, ab129184 (RRID: AB_11150422), dil. 1:500
 anti-CTCF, Active Motif, 61311 (RRID: AB_2614975), dil. 1:500
 anti-SON, Rockland Immunochemicals, ABIN768615 (RRID: N/A), dil. 1:500
 anti-SON, Merck Millipore, HPA031755 (RRID: N/A), dil. 1:500
 anti-GFP, Abcam, ab290 (RRID: AB_303395), dil. 1:1000
 anti-HMGB1/2, Abcam, ab190377 [1F3] (RRID: AB_3711424), dil. 1:500
 anti-HMGB2, Abcam, ab67282 (RRID: AB_1140885), dil. 1:500
 anti-SC35, Novus Biologicals, NB100-1774 (RRID: AB_10128431), dil. 1:500
 anti-SQSTM1, Cell Signaling, 88588 (RRID: AB_2800125), dil. 1:500
 anti-SRRM2, Thermo Fisher, PA5-59559 (RRID: AB_2647934), dil. 1:500
 anti-ATXN2, Santa Cruz, sc-515602 (RRID: N/A), dil. 1:500
 anti-FMR1, Santa Cruz, sc-293156 (RRID: N/A), dil. 1:500

Secondary:

anti-mouse Cy3, Abcam, ab97035 (RRID: AB_10680176), dil. 1:1000
 anti-rabbit Alexa488, Abcam, ab150077 (RRID: AB_2630356), dil. 1:1000
 anti-rabbit-Cy3, Abcam ab6939 (RRID: AB_955021), dil. 1:1000
 anti-mouse-A647, Abcam ab150115 (RRID: AB_2687948), dil. 1:1000
 anti-rabbit-A647, Abcam ab150083 (RRID: AB_2714032), dil. 1:1000
 anti-rat-A647, Abcam ab150155 (RRID: AB_2813835), dil. 1:1000

Secondary for STED imaging:

anti-mouse IgG-Abberior® STAR 580, Sigma-Aldrich, 52403, dil. 1:1000
 anti-rabbit IgG-Abberior® STAR 580, Sigma-Aldrich, 41348, dil. 1:1000

Validation

All antibodies come with specific validation information from their respective manufacturer, but we have also tested the key ones via blotting when performing siRNA-mediated knockdowns (i.e., those against CTCF, HMGB2, BANF1, and SRRM2).

Eukaryotic cell lines

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

Cell line source(s)

We have used the most popular cell model for replicative senescence, female fetal lung fibroblasts (IMR90), which are directly purchased and validated from the Coriell Repository and match those used by the ENCODE project. Similarly, our human umbilical vein endothelial cells (HUVECs) we purchased as a FACS-marker-validated pool from Lonza Bioscience. We also performed a single IF experiment in low-passage GM00038 (6-year-old donor) and AG05247 skin fibroblast cells (87-year-old donor; also from Coriell).

Authentication

Authentication and detailed documentation for each cell line is provided by the Coriell Repository and Lonza on purchase.

Mycoplasma contamination

All cell lines in our institute are tested for mycoplasma every 6 months; our IMR90 and HUVECs stocks have never tested positive for mycoplasma since 2018 that we started using them in Goettingen.

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

IMR90s and HUVECs do not belong to the commonly misidentified lines of the ICLAC register.

Plants

Seed stocks

N/A

Novel plant genotypes

N/A

Authentication

N/A

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.

All ChIPmentation data generated in this study have been deposited to the NCBI Gene Expression Omnibus (GEO) and are available under accession number GSE269858

Files in database submission

Raw .fastq files, plus mapped .bam files and .BED peak lists (also provided as Supplementary Data 6 with the manuscript)

Genome browser session
(e.g. [UCSC](#))

N/A

Methodology

Replicates

Each ChIPmentation experiment was performed in at least two independent replicates

Sequencing depth

Each replicate was sequenced to ~50 million 75-nt reads on an Illumina NextSeq platform

Antibodies

anti-HMGB2, Abcam, ab190377-1F3 (RRID: N/A); 2 ug per IP was used.
anti-SON, Merck Millipore, HPA031755 (RRID: N/A); 2 ug per IP was used.

Peak calling parameters

ChIPmentation data were mapped and quality controled with the NGseqBasic pipeline (<https://github.com/Hughes-Genome-Group/NGseqBasic>; hg38 genome assembly) using its default settings.

Data quality

Peak lists are provided in Supplementary Data 6 and are 6435, 25806, and 5923 for SON in senescent, SON in proliferating, and HMGB2 in proliferating IMR90, respectively. These peaks are satisfy a 1% FDR threshold and a fold-enrichment of >2.5-fold.

Software

After sequence alignment and deduplication, bigWig coverage tracks were generated with the deepTools (ver 3.3.0) bamCoverage function using a binning size of 10bp. ChIPmentation coverage tracks were normalized using RPKM. Coverage tracks of replicates were merged using UCSC bigWigMerge (ver 2.0).