

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	Microscopy data was collected with Micromanager
Data analysis	All data analysis was done with Matlab 2021a, and 2024b and is freely available at: https://zenodo.org/records/13351421 . The image processing software, CellDisect, used for cell and nuclear segmentation, was previously published in reference 50. The spot detection software, TrueSpot, has been submitted for publication and is available through the following Zenodo link: https://zenodo.org/records/13345321

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](#)

A data availability statement is provided in the manuscript.

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	N/A
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	Sample size for replica was determined by reproducing the results. Number of cells were determined to have sufficient numbers of cell in each bin. Each sample consists of hundreds of cells.
Data exclusions	No data was excluded as outlined in the manuscript.
Replication	Biological replica were determined by producing reducible results as outlined in the manuscript.
Randomization	For each replica data set, a randomized sample was produced for statistical testing as outlined in the manuscript.
Blinding	Blinding was not relevant because we compared our results against a randomized data set.

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	1) Primary antibody: Antigen: H3K36me3, Host / Isotype: Rabbit; Coupled Fluorophore: none, Manufacturer: Cell Signaling, Catalog Number: 4909S, Dilution: 1:270; 2) Secondary Antibody: Antigen: Anti-Rabbit, Host / Isotype: Goat; Coupled Fluorophore: Alexa Fluor® 488, Manufacturer: Cell Signaling, Catalog Number: 4412S, Dilution: 1:2000;
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Validation

The primary antibody was diluted as described in the manuscript.
The secondary antibody gave no signal without primary antibody.

Eukaryotic cell lines

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

Cell line source(s)

F1 2-1 mESCs (Luikenhuis et al., 2001) were obtained from the lab of Dr. Rudolf Jaenisch

Authentication

Validated by karyotyping.

Mycoplasma contamination

Cell lines were not tested for mycoplasma contamination. But we have verified Jurkat cell that are not mycoplasma contaminated and that have been grown in the same cell culture facility.

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

N/A

Plants

Seed stocks

N/A

Novel plant genotypes

N/A

Authentication

N/A