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Corresponding author(s): Luke D. Lavis

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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	The Shimadzu analytical HPLC instrument was controlled by Shimadzu LabSolutions 5.51 software. The Shimadzu preparative HPLC instruments were controlled by Shimadzu LabSolutions 5.42 SP4, 5.85, or 5.97 software. The Shimadzu LC/MS system was controlled by LabSolutions 5.42 SP4 software. The Agilent LC/MS system was controlled by Agilent LC/MSD ChemStation Rev. B04.02 [54] software. The Biotage Isolera flash chromatography system was controlled by Biotage OS 852M software. The Buchi Rotavapor was running Buchi firmware version 01.02.00.00 with a vacuum pump running Buchi firmware version 01.00.00.00 and controller running Buchi firmware 01.01.00.00. The NMR was controlled by IconNMR 4.7.7 Build 20 software. The Cary Model 100 spectrometer was controlled by CaryWinUV 4.20(468) software. The Cary Eclipse was controlled by Cary Eclipse Scan Application 1.1(132) software. The Quantaurus Quantum Yield spectrophotometer was controlled by PLQY software U6039-05 3.4.2. The Zeiss LSM 880 confocal microscope was controlled by Zen2.1 SP3 software. The Leica SP8 Falcon confocal microscope was controlled by Leica Application Suite X 3.5.6.21594 software. The Nikon Eclipse Ti microscope was controlled by NIS-Elements AR 4.40.0, 64-bit (build 1084) software. The TissueFAXS 200 confocal slide scanner was controlled by TissueFAXS 5.3.6245.1016 software. The CytoFLEX flow cytometer was controlled with CytoFLEX CytExpert v.2.3 software.
Data analysis	NMR data were analyzed using Bruker TopSpin 3.2 or MestReNova 14.1.1-24571 software. Spectra and other graphical data were analyzed using GraphPad Prism 8 for macOS version 8.4.2 (464) software. Fluorescence microscopy images were processed and analyzed using the following software: Zen2.1 SP3 software from Zeiss, NIS-Elements AR 4.40.0, 64-bit (build 1084) software from Nikon, or FIJI Version:2.0.0-rc-69/1.52p; Build:269a0ad53f. Flow cytometry data was analyzed using FlowJo v.10.6.1 software.

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

The data that support the findings of this study are provided in the Source Data files or available from the corresponding author upon request.

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	All quantitative imaging experiments were performed on replicate samples or fields of view as reported in the Methods and Figure legends.
Data exclusions	No data were excluded from this study.
Replication	All imaging and flow cytometry data were repeated and showed similar results.
Randomization	We compared a relatively small number of compounds allowing the use of cell culture samples that were the same age, treated in the same way, and analyzed by a standard protocol. Randomization was not necessary for these experiments.
Blinding	We compared a relatively small number of compounds allowing the use of cell culture samples that were the same age, treated in the same way, and analyzed by a standard protocol. Blinding was not necessary for these experiments.

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

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	U2OS cells were from ATCC. The mouse ES JM8.N4 cells, derived from the C57BL/6N mouse strain, were a gift from Robert Tjian's lab (University of California, Berkeley).
Authentication	The U2OS cells were authenticated by ATCC and were not re-authenticated at Janelia. The JM8.N4 cells were authenticated by short tandem repeat DNA profiling and approved by the NIH 4D Nucleome project as a Tier2 cell line.
Mycoplasma contamination	The Janelia Cell Culture Facility regularly tests for mycoplasma contamination; these cell lines 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	Adult C57/BL6 male mice, 2-4 months old, were used to express a GFP-HaloTag fusion protein throughout the brain.
Wild animals	No wild animals were used in this study.
Field-collected samples	No field-collected samples were used in this study.
Ethics oversight	All experimental protocols were conducted according to the National Institutes of Health guidelines for animal research and were approved by the Institutional Animal Care and Use Committee at the Janelia Research Campus, HHMI.

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

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	Wild type mouse embryonic stem (ES) cells or ES cells stably expressing the SNAP-tag–histone H2B were plated into onto flat-bottom 96-well plates precoated with 0.1% gelatin (Corning). Cells at 70% confluency were stained with JF549–cpSNAP-tag ligand (2STL) or JF552–cpSNAP-tag ligand (13STL) at different concentrations (3 nM, 10 nM, 30 nM, 100 nM, 300 nM) for 15 min or at different time points (15 min, 30 min, 60 min, 120 min, 210 min) at 10 nM concentration. Cells were washed 3× for 10 min, trypsinized, and loaded onto the flow cytometer equipped with a plate loader.
Instrument	CytoFLEX S flow cytometer equipped with a plate loader (Beckman Coulter).
Software	Data was collected with the CytoFLEX CytExpert v.2.3 operating software and analyzed with FlowJo v.10.6.1 software.
Cell population abundance	The cells were not sorted. For the experimental replicates, the mouse embryonic stem cell line used for flow cytometry analysis had between 85–98% expression of SNAP-tag–histone H2B as determined against negative fluorescence gating.
Gating strategy	The mouse ES cell population was designated based on its forward light scatter (FSC) and side light scatter (SSC) characteristics (Supplementary Fig. 3a). The "nonfluorescent" cell population was determined from control ES cell samples not incubated with SNAP-tag ligands, plotting SSC-H vs. PE fluorescence (phycoerythrin; 585 nm with a 42 nm bandpass; Supplementary Fig. 3b). Sample dilution and flow rate were adjusted to optimize event recordings for the 96-well microplate format. The settings were as follows: FSC avalanche photodiode (AP) detector gain setting = 12; SSC AP detector gain setting = 130; FSC threshold Automatic; PE channel AP gain setting = 1.

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