nature portfolio

Corresponding author(s): Benjamin Deneen

Last updated by author(s): Sep 19, 2025

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 (*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	Confocal imaging was conducted with Zeiss LSM 880 laser scanning confocal microscope and Zen software (v3.1). Calcium imaging: GECIquant (v1.0), ImageJ (v1.54f), and Clampfit (v10.7) were used to analyze calcium signals. Electrical signals were digitized and sampled at 50 µs intervals with Digidata 1550B and Multiclamp 700B amplifier (Molecular Devices, CA, USA) using pCLAMP (v10.7) software. FACS-sorting was performed using a Becton Dickinson FACSaria II with FACSDiva software. RNA-seq was performed on Illumina Nextseq system suite. ChIP-seq was performed on Illumina Nextseq system suite Images in schematics were created using Biorender.com.
Data analysis	ImageJ (v1.54f) was used for confocal image analysis. Clampfit (10.7) was used for electrophysiological data analysis. The following were used for calcium image analysis: GECIquant, ImageJ (v1.54f), Clampfit (10.7). The following were used for RNA-seq and bioinformatic analyses: RNA-seq data was processed using fastQC (v0.11.7), MultiQC (v1.6), STAR (v2.5.0a), HOMER (v4.10), and DESeq2 (v1.30.1). The follwing were used for ChIP-seq and bioinformatic analysis: ChIP-seq data was processed using fastQC (v0.11.7), MultiQC (v1.6), bowtie2 (v 2.2.6), and HOMER (v4.10) software suite. Integrated Genome Browser-compatible files were made using samtools (v1.9), sort and index, deepTools (v3.2.0), and bamCompare (v3.2.0). Graphpad Prism (v10) was used for all statistical analysis.

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

All primary antibodies used in this study are listed in the Methods. RNA-seq data were deposited in the GEO database under the accession number (ChIP: GSE294933 and RNAseq: GSE294900). Source data are provided in this paper.

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	All of the information in the AD groups (n=4) and age-matched control (n=4) is in Extended Data Fig. 4e.
Reporting on race, ethnicity, or other socially relevant groupings	N/A
Population characteristics	N/A
Recruitment	N/A
Ethics oversight	The BCM IRB and the BCM determined that this study does not constitute human subjects research.

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 formal estimates of statistical power were done. Sample sizes were based on previous publications using similar techniques. Animal numbers were minimized to conform to ethical guidelines while maintaining adequate numbers to derive accurate measurements.
Data exclusions	There were no data exclusions.
Replication	All attempts at replication were successful. Mice from multiple litters were used to attain at least 3 biological replicates per group for each measurement.
Randomization	Mice were randomly allocated to groups when possible. Where dependent on genotype, mice from multiple litters were randomly selected to be used.
Blinding	Investigators were blinded to group allocation during experimentation and 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

The following primary antibodies were used: chicken anti-GFP (1:1000, Abcam, ab13970), chicken anti-GFAP (1:1000, Abcam, ab4674), mouse anti-S100B (1:500, Invitrogen, MA1-25005), mouse anti-Cathepsin D (1:200, R&D system, AF1029), Rat anti-CD68 (1:200, BIORAD, MCA1957), mouse anti-NeuN (1:500, Sigma-Aldrich, MAB377), rabbit anti-NeuN (1:100, Sigma-Aldrich, ABN78), rabbit anti-Iba1 (1:1000, FuJIFILM Wako,019-19741), goat anti-Iba1 (1:500, abcam, ab5076), guinea pig anti-vGlut1(1:1000, Sigma-Aldrich, AB5905), mouse anti-PSD95 (1:500, Invitrogen, MA1-046), rabbit anti-PSD95 (1:500, Invitrogen, 51-6900), mouse anti-Aβ (1:1000, Abcam, ab126649), rabbit anti-Aβ (1:1000, Abcam, ab201060), rat-anti-Lamp1 (1:1000, DSHB, 1D4B), rabbit anti-Sox9 (1:1000, Abcam no. ab5535), goat-anti-Sox10 (1:500, Santacruz, sc-17342), rabbit anti-LRP1 (1:200, ThermoFisher #37-7600), rabbit anti-CD36 (1:50, Thermofisher #PA1-16813), rabbit anti-RAGE (1:500, Thermofisher #PA1-075) and rabbit anti-Megf10 (1:200, Milipore sigma, ABC10).
The following secondary antibodies were used:goat anti-mouse Alexa Fluor 568 (Thermo Fisher no. A11004), goat anti-mouse Alexa Fluor 647 (Thermo Fisher no. A21235), goat anti-chicken Alexa Fluor 488 (Thermo Fisher no. A11039), goat anti-rabbit Alexa Fluor 568 (Thermo Fisher no. A11036) , goat anti-rabbit Alexa Fluor 647 (Thermo Fisher no. A21244), goat anti-Rat Alexa Fluor 568 (Thermo Fisher no. A11077) and goat anti-Rat Alexa Fluor 647 (Thermo Fisher no.A21247)

Validation

Antibodies were validated by manufacturers and used in previously published work. Appropriate primary antibody dilutions were determined by testing multiple dilutions on sample mouse brain tissue. Secondary antibody only controls were used to confirm lack of signal in the absence of primary antibodies.
1. chicken anti-GFP (1:1000, Abcam, ab13970): A polyclonal chicken antibody, supplied by Abcam, raised against Green fluorescent protein (Water jellyfish), cited in 4979 publications, with 214 published images. Applications used include IHC, IHC-IF, IF, ICC-IF, and 20 others. Recent publication PMID: 39294371
2. chicken anti-GFAP (1:1000, Abcam, ab4674): A polyclonal chicken antibody, supplied by Abcam, raised against Glial fibrillary acidic protein (Human), cited in 930 publications, with 30 published images. Applications used include IHC, IHC-IF, ICC, ICC-IF, and 8 others. Recent publication PMID: 39231983
3. mouse anti-Aβ (1:1000, Abcam, ab126649): A monoclonal mouse antibody, supplied by Abcam, raised against Amyloid-beta precursor protein (Human), cited in 39 publications, with 5 published images. Applications used include IHC, IHC-IF, ICC-IF, and WB. Recent publication PMID: 36721205
4. rabbit anti-Aβ (1:1000, Abcam, ab201060): A recombinant monoclonal rabbit antibody, supplied by Abcam, raised against Amyloid-beta precursor protein (Human), cited in 121 publications, with 3 published images. Applications used include WB, IHC, IHC-IF, IF, and 1 other. Recent publication PMID: 38548872
5. rat-anti-Lamp1 (1:1000, DSHB, 1D4B): A monoclonal rat antibody, supplied by Developmental Studies Hybridoma Bank, raised against Lysosome-associated membrane glycoprotein 1 (Mouse), cited in 734 publications, with 38 published images. Applications used include WB, ICC-IF, IF, IHC, and 7 others. Recent publication PMID: 38802590
6. rabbit anti-Sox9 (1:1000, MiliporeSigma no. ab5535):A polyclonal rabbit antibody, supplied by MilliporeSigma, raised against Transcription factor SOX-9 (Human), cited in 1325 publications, with 40 published images. Applications used include IHC, IHC-IF, WB, IF, and 13 others. Recent publication PMID: 39284798
7.rabbit anti-LRP1 (1:200, ThermoFisher #37-7600): A monoclonal mouse antibody, supplied by Invitrogen Antibodies, raised against Prolow-density lipoprotein receptor-related protein 1 (Human), cited in 11 publications, with 1 published image. Applications used include WB, IP, and IHC. Recent publication PMID: 35835845
8. rabbit anti-CD36 (1:50, Thermofisher #PA1-16813): A polyclonal rabbit antibody, supplied by Invitrogen Antibodies, raised against Platelet glycoprotein 4 (Human), cited in 21 publications, with 1 published image. Applications used include WB, IHC, ICC-IF, IHC-IF, and 2 others. Recent publication PMID: 33692340
9.rabbit anti-RAGE (1:500, Thermofisher #PA1-075): A polyclonal rabbit antibody, supplied by Invitrogen Antibodies, raised against MAPK/MAK/MRK overlapping kinase (Human), cited in 18 publications, with 2 published images. Applications used include WB, IHC, IF, IHC-Frozen, and 3 others. Recent publication PMID: 31988531
10. rabbit anti-Megf10(1:200, Milipore sigma, ABC10): A polyclonal rabbit antibody, supplied by MilliporeSigma, raised against Multiple epidermal growth factor-like domains protein 10 (Mouse), cited in 10 publications. Applications used include WB and IHC.Recent publication PMID: 35228547
11. mouse anti-S100B (1:500, Invitrogen, MA1-25005): A monoclonal mouse antibody, supplied by Invitrogen, raised against S100B, cited in 7 publications. Applications used included WB and IHC. Recent publication PMCID: PMC11212328
12. mouse anti-Cathepsin D (1:200, R&D system, AF1029): A polyclonal goat antibody, supplied by R&D Systems, raised against Cathepsin D (Mouse), cited in 77 publications, with 2 published images. Applications used include WB, IHC, ICC, ICC-IF, and 6 others. Recent publication PMCID: PMC11965215
13. Rat anti-CD68 (1:200, BIORAD, MCA1957): A monoclonal rat antibody, supplied by Bio-Rad (Formerly AbD Serotec), raised against Macrosialin (Mouse), cited in 1302 publications, with 32 published images. Applications used include IHC, IHC-IF, IHC-Frozen, IF, and 14 others. Recent publication PMCID: PMC11946907
14. mouse anti-NeuN (1:500, Sigma-Aldrich, MAB377): A monoclonal mouse antibody, supplied by MilliporeSigma, raised against RNA binding protein fox-1 homolog 3 (Mouse), cited in 6814 publications, with 280 published images. Applications used include IHC, IHC-IF, IF, ICC, and 16 others. Recent publication PMCID: PMC11978576

15. rabbit anti-NeuN (1:100, Sigma-Aldrich, ABN78): A polyclonal rabbit antibody, supplied by MilliporeSigma, raised against RNA binding protein fox-1 homolog 3 (Mouse), cited in 746 publications, with 24 published images. Applications used include IHC, IHC-IF, IF, WB, and 7 others. Recent publication PMCID: PMC11814324
16. rabbit anti-Iba1 (1:1000, FuJIFILM Wako,019-19741): An antibody, supplied by FUJIFILM Cellular Dynamics, Inc., cited in 688 publications, with 3 published images. Applications used include IHC, IHC-IF, ICC, IF, and 3 others. Recent publication PMCID: PMC11909085
17. goat anti-Iba1 (1:500, abcam, ab5076): A polyclonal goat antibody, supplied by Abcam, raised against Allograft inflammatory factor 1 (Human), cited in 1853 publications, with 159 published images. Applications used include IHC, IHC-IF, WB, IF, and 11 others. Recent publication PMCID: PMC11976812
18. guinea pig anti-vGlut1(1:1000, Sigma-Aldrich, AB5905): A polyclonal guinea pig antibody, supplied by MilliporeSigma, raised against Vesicular glutamate transporter 1 (Rat), cited in 734 publications, with 15 published images. Applications used include IHC, ICC, WB, IHC-IF, and 7 others. Recent publication PMCID: PMC11906744
19. mouse anti-PSD95 (1:500, Invitrogen, MA1-046): A monoclonal mouse antibody, supplied by Invitrogen Antibodies, raised against Disks large homolog 4 (Rat), cited in 403 publications, with 63 published images. Applications used include WB, ICC, IHC, ICC-IF, and 11 others. Recent publication PMCID: PMC11821914
20. rabbit anti-PSD95 (1:500, Invitrogen, 51-6900): A polyclonal rabbit antibody, supplied by Invitrogen Antibodies, raised against Disks large homolog 4 (Rat), cited in 183 publications, with 12 published images. Applications used include IHC, IHC-IF, WB, IF, and 9 others. Recent publication PMCID: PMC11531546
21. goat-anti-Sox10 (1:500, Santacruz, sc-17342): A polyclonal goat antibody, supplied by Santa Cruz Biotechnology, raised against Transcription factor SOX-10 (Human), cited in 76 publications. Applications used include WB, IHC, IHC-IF, FC/FACS, and 2 others. Recent publication PMCID: PMC10966798

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals	The following transgenic mouse strains were used: APPNLGF(EIKEN center for Brain Science), Rosa-CAG-LSL-tdTomato (Ai14; Jackson Laboratory,007914), Aldh1l1-CreER (Jackson Laboratory, 029655), Aldh1l1-GFP (RRID:MMRRC_011015-UCD), and Sox9-flox. All mice were on a C57/b6 background.
Wild animals	No wild animals were used in this study.
Reporting on sex	Mice of both sexes were used. Data was not stratified nor analyzed based on sex.
Field-collected samples	No field-collected samples were used.
Ethics oversight	Experiments were conducted in accordance with protocols approved by the Institutional Animal Care and Use Committee at Baylor College of Medicine.

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

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 sequencing data will be made available through the NCBI Gene Expression Omnibus (GEO) Repository and will be publicly available upon publication. Early access code will be given upon reviewer request.
Files in database submission	FASTQ and bigWig data for all ChIP-seq samples performed in this study
Genome browser session (e.g. UCSC)	UCSC Genome Browser assembly ID: mm10

Methodology

Replicates	Each group was collected from 6 mice
Sequencing depth	All ChIP-seq were done with single-end sequencing and have > 95% of overall alignment rate and > 64% of the uniquely mapped reads.Total read counts range from 24-48 million reads.
Antibodies	Rabbit anti-Sox9 (1:1000, MiliporeSigma no. ab5535)
Peak calling parameters	The find peaks command in factor mode was used to filter ChIP peaks enriched over input control using by the HOMER (v4.10).
Data quality	Fastq files were validated by MultiQC (V1.6). Peaks were filtered using factor and findPeaks command in HOMER suite (v4.10) as described above.
Software	Sequencing files were processed and analyzed ChIP-seq data were analyzed using fastQC (v0.11.7), MultiQC (v1.6), bowtie2 (v 2.2.6), and HOMER (v4.10) software suite. Integrated Genome Browser-compatible files were made using samtools (v1.9), sort and index, deepTools (v3.2.0), and bamCompare (v3.2.0).

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	Mice were perfused with cold saline and hippocampi were dissected on ice. Dissected tissue was dissociated as previously described (PMID: 28166219).
Instrument	BD FACSAria II
Software	BD FACSDiva software
Cell population abundance	Enrichment of astrocytes and de-enrichement of neurons were confirmed by qPCR of cDNA ibraries prepared from sorted samples.
Gating strategy	Singlet gating was done based on forward and side scatter. Singlet events were assessed for GFP and mCherry fluorescence. Non-fluorescent brain tissue was used as a negative control to set the double negative population gate. We also performed single color controls to establish ideal voltage, potential compensation, and gating. The GPF signal was detected with a 488nm laser, through a 505nm long pass filter, and 530/30 detector. mCherry signal was detected with 561nm laser, through a 570 long pass filter, and 585/51 detector.

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