nature portfolio

Corresponding author(s): Ulrich Muller

Last updated by author(s): Aug 14, 2024

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	Zen 2.3 Microscopy Software; Illumina NovaSeq 6000; Illumina's BaseSpace software.
Data analysis	Imaris versions 8 to10 (Bitplane - OXFORD Instruments); ImageJ (Fiji), version 2.14.0/1.54f (National Institutes of Health); GraphPad Prism 10 (Dotmatics); Adobe Photoshop version 25.4 (Adobe); Adobe Illustrator version 28(Adobe); Snapgene version 5.0.8 (Biotech LLC); Cell Ranger (10X Genomics); R Project for Statistical Computing (The R foundation); Python Programming Language (Python Software Foundation); Enrichr version 3.2 (Mount Sinai Center for Bioinformatics); Ensembl genome browser 111. The code for the scRNAsequencing analysis is available on: DOI: 10.5281/zenodo.12582809 https://zenodo.org/doi/10.5281/zenodo.12582808

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

The raw single cell RNA sequencing datasets generated in this study have been deposited in the GEO repository database under the accession code GSE229129 (https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE229129). Raw single cell RNA sequencing datasets for the monkey data (Zhu, X. et al.Sci Adv 8, 1–17 (2022)) are deposited in the SRA repository database under the accession code SRP366952 (https://www.ncbi.nlm.nih.gov/sra/?term=SRP366952). All the other data generated in this study are provided in the Supplementary Information and Supplementary Data files. Source Data are provided with 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	*Use the terms sex (biological attribute) and gender (shaped by social and cultural circumstances) carefully in order to avoid confusing both terms. Indicate if findings apply to only one sex or gender; describe whether sex and gender were considered in study design; whether sex and/or gender was determined based on self-reporting or assigned and methods used. Provide in the source data disaggregated sex and gender data, where this information has been collected, and if consent has been obtained for sharing of individual-level data; provide overall numbers in this Reporting Summary. Please state if this information has not been collected. Report sex- and gender-based analyses where performed, justify reasons for lack of sex- and gender-based analysis.*
Reporting on race, ethnicity, or other socially relevant groupings	*Please specify the socially constructed or socially relevant categorization variable(s) used in your manuscript and explain why they were used. Please note that such variables should not be used as proxies for other socially constructed/relevant variables (for example, race or ethnicity should not be used as a proxy for socioeconomic status). Provide clear definitions of the relevant terms used, how they were provided (by the participants/respondents, the researchers, or third parties), and the method(s) used to classify people into the different categories (e.g. self-report, census or administrative data, social media data, etc.) Please provide details about how you controlled for confounding variables in your analyses.*
Population characteristics	*Describe the covariate-relevant population characteristics of the human research participants (e.g. age, genotypic information, past and current diagnosis and treatment categories). If you filled out the behavioural & social sciences study design questions and have nothing to add here, write "See above."*
Recruitment	*Describe how participants were recruited. Outline any potential self-selection bias or other biases that may be present and how these are likely to impact results.*
Ethics oversight	*Identify the organization(s) that approved the study protocol.*

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 predetermination of sample size was calculated. We defined a minimum of n=3 and a maximum of n=10 animals to be sufficient based on the internal controls and degree of variability between samples in the different experiments.
Data exclusions	No data were excluded from the imaging analysis. For the single-cell RNA sequencing analysis, although we started with seven Brn1/2-cKO and seven wild-type littermates of both sexes (Supplementary Fig. 5a), the genotype for one of the wild-type samples could not be confirmed and therefore this sample was removed from the analysis (Fig. 1f). Additionally, for the transcriptional waves along pseudotime projection analysis, Batch 1 failed to show temporal difference in the latent space, we removed this batch from the following pseudotime analysis. Subsequently, we have confirmed that the control and Brn1/2-cKO cells from Batch 1 showed similar average age-related expression pattern compared with the other batches within the same genotype for the vast majority of the genes used to define the transcriptional waves.
Replication	At least 3 individual animals were analyzed per genotype or experimental condition to ensure data reproducibility.

Randomization	Sample collection was randomized to avoid any bias in the analyzes.
Blinding	The investigators were blinded to group allocation during imaging data collection. Sample collection for single cell RNA sequencing was not performed blindly since precise identification and isolation of cells is essential for accurate experimental design.

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

anti-L1 (rat monoclonal, MAB5272 Millipore Sigma, clone 324, current lot: 3469466)
anti-TLE4 (rabbit polyclonal, ab64833 Abcam, current lot: GR3341430-7)
anti-CTIP2 (rat monoclonal, ab18465 Abcam, clone 25B6, current lot: GR322373-4)
anti-RORb (mouse monoclonal, PP-N7927-00 R&D systems, clone N7927, current lot: A-2)
anti-CUX1 (rabbit polyclonal, 11733-1-AP Proteintech, current lot: 00050220)
anti-GFP (chicken polyclonal, GFP-1020 Aves Labs Inc., current lot: GFP917979)
anti-BRN2 (rabbit polyclonal, PA5-30124 Thermo Fisher Scientific, current lot: UB2712455B)
anti-BRN2 (rabbit monoclonal, 12137 Cell Signaling, clone D2C1L)
anti-BRN1 (rabbit polyclonal, sc-6028-R Santa Cruz Biotechnology, lot: J2512);
anti-NeuN (mouse monoclonal, MAB377 Millipore Sigma, clone A60, current lot: 3233107);
anti-Ki67 (rabbit polyclonal, ab15580 Abcam, current lot: GR3285096-1)
anti-Ki67 (rat monoclonal, 14-5698-82 Thermo Fisher Scientific, clone SolA15, current lot: 2496198)
anti-RFP (rat monoclonal, 5F8 ChromoTek, clone 5F8, current lot: 90228002AB-05)
anti-RFP (rabbit polyclonal, ab62341 Abcam, current lot: 1053546-1)
anti-Pax6 (mouse monoclonal, MA1-109 Thermo Fisher Scientific, clone 13B10-1A10, current lot: XD341788)
anti-TBR2 (rat monoclonal, 14-4875-82 Thermo Fisher Scientific, clone Dan11mag, current lot: 2488481)
anti-TBR2 (rabbit polyclonal, ab183991 Abcam, clone EPR19012, current lot: GR3237130-1)
anti-NEUROD2 (rabbit polyclonal, ab104430 Abcam, current lot: GR3414328-2)
anti-HES1 (rabbit polyclonal, R. Kageyama gift, Shimojo, H., Ohtsuka, T. & Kageyama, R. Oscillations in Notch Signaling Regulate Maintenance of Neural Progenitors. Neuron 58, 52–64 (2008))
anti-pVIM (mouse monoclonal, D076-3 MBL, clone 4A4, current lot: 038)
anti-OLIG2 (rabbit polyclonal, AB9610 Millipore Sigma, current lot: 2728398)
anti-SOX9 (1:1000, rabbit monoclonal, ab185966 Abcam, clone EPR14335-78 current lot: GR3194558-4)
anti-GFAP (1:1000, chicken polyclonal, AB5541 Millipore Sigma, current lot: 3489761)
anti-PH3 (1:1000, rat monoclonal, ab10543 Abcam, clone HTA28, current lot: 1052214-1)
anti-g-TUBULIN (1:500, goat polyclonal, A. Holland gift, Levine, M. S. et al. Centrosome Amplification Is Sufficient to Promote Spontaneous Tumorigenesis in Mammals. Dev Cell 40, 313-322.e5 (2017))
anti-Centrin (1:500, rabbit polyclonal, A. Holland gift, Moyer, T. C. & Holland, A. J. PLK4 promotes centriole duplication by phosphorylating STIL to link the procentriole cartwheel to the microtubule wall. Elife 8, e46054 (2019)).
anti-NRP1 (1:500, goat polyclonal, AF566 R&D systems, current lot:ETH0915031)
anti-CRE (1:200, mouse monoclonal, MAB3120 Millipore Sigma, clone 2D8, current lot: 3213901)

Validation

Anti-L1 (validation manufacturer's statement: IC, IH, WB)
Anti-TLE4 (validated by IHC in E16.5 mouse brain sections in the manufacturer's website);
Anti-CTIP2 (validated by IHC in mouse brain sections at different ages in the manufacturer's website);
Anti-RORb (validated in this study by IHC and specific labeling of layer IV in mouse brain sections);
Anti-CUX1 (validated by IHC in adult mouse brain sections in the manufacturer's website);
Anti-GFP (validated by IHC in GFP transgenic mouse brain sections in the manufacturer's website);
Anti-BRN2 (validated by IHC in mouse and rat brain sections in the manufacturer's website; validated in this study by IHC in brain sections from control and Brn1/2 knockout mice);
Anti-BRN1 (validated in this study by IHC in brain sections from control and Brn1/2 knockout mice);
Anti-NeuN (validated by IHC in mouse brain sections in the manufacturer's website);
Anti-Ki67 (validated by IHC in mouse brain sections in the manufacturer's websites);
Anti-RFP (validated by IHC in dsRed transgenic mouse brain sections in the manufacturer's website and their referred references);
Anti-Pax6 (validation manufacturer's statement: This Antibody was verified by relative expression to ensure that the antibody binds

to the antigen stated);
Anti-TBR2 (validated by IHC in mouse brain sections at different ages in the manufacturer's websites);
Anti-NEUROD2 (validated by IHC in mouse brain tissue in the manufacturer's website);
Anti-HES1 (validated by R. Kageyama's group: Shimojo, H., Ohtsuka, T. & Kageyama, R. Oscillations in Notch Signaling Regulate Maintenance of Neural Progenitors. Neuron 58, 52–64 (2008));
Anti-pVIM (manufacturer's referred references);
Anti-OLIG2 (validation manufacturer's statement: tested by IHC on human, rat and mouse brain tissues);
Anti-SOX9 (validated by IHC in mouse brain tissue in the manufacturer's website);
Anti-GFAP (validation manufacturer's statement: The antibody stains processes of astrocytes in sections of brain tissues);
Anti-PH3 (manufacturer's referred references);
Anti-g-TUBULIN (validated by A. Holland's group: Levine, M. S. et al. Centrosome Amplification Is Sufficient to Promote Spontaneous Tumorigenesis in Mammals. Dev Cell 40, 313-322.e5 (2017));
Anti-Centrin (validated by A. Holland's group: Moyer, T. C. & Holland, A. J. PLK4 promotes centriole duplication by phosphorylating STIL to link the procentriole cartwheel to the microtubule wall. Elife 8, e46054 (2019)).
Anti-NRP1 (validated by IHC in mouse brain tissue in the manufacturer's website)
Anti-CRE (validated by IHC in mouse brain tissue in the manufacturer's website)

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	293 [HEK-293] - CRL-1573 ™ cells were obtained from ATCC
Authentication	No authentication was performed
Mycoplasma contamination	Cells tested negative for mycoplasma
Commonly misidentified lines (See ICLAC register)	N/A

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	Emx1-Cre [B6.129S2-Emx1tm1(cre)Krj], Brn2 conditional knockout [Brn-2fl/fl] and Brn1 conditional knockout [Brn-1fl/fl - generated in this study] mice were crossed at adult age to obtain Emx1-Cre;Brn1fl/fl;Brn2fl/fl mice and control littermates at the different ages reported in this study (Embryonic age 12.5 (E12.5) to postnatal age 13 (P13)). Time-pregnancy crossings of Brn2 conditional knockout [Brn-2fl/fl] and Brn1 conditional knockout [Brn-1fl/fl - generated in this study] adult mice were used for in utero electroportation experiments at E12.5 and E14.5 (collection at P13 or at E16.5). Time-pregnant C57BL/6J wild type females were used to collect E11.5 wild-type embryos. Time-pregnant wild-type ferrets were obtained from Marshall BioResources for in utero electroportation experiments at E35 (collection at E37).
Wild animals	This study did not involve wild animals.
Reporting on sex	Since no differences were found between males and females for the phenotypes described in this study both sexes were included in the analysis. For embryonic samples we only determined sex for the scRNAsequencing experiments in mice: E12.5 control = 2 males and 1 female; E12.5 Brn1/2-cKO = 2 males and 1 female; E14.5 control = 2 males and 1 female; E14.5 Brn1/2-cKO = 1 males and 3 females. No sex was determined for the Ferret analysis and P0 analysis in mice. For P13 analysis in mice we used: Fig. 1b and 1c: control = 4 males and 1 female; Brn1/2-cKO = 4 males and 2 females; Supplementary Fig. 2g: TLE4: control = 3 males and 1 female, Brn1/2-cKO = 3 males and 1 female; CTIP2: control = 4 males and 1 female, Brn1/2-cKO = 3 males and 2 females; RORb: control = 3 males, Brn1/2-cKO = 3 males and 1 female; CUX1: control = 4 males, Brn1/2-cKO = 4 males and 2 females; Supplementary Fig. 3a': control = 3 males and 2 females, Brn1/2-cKO = 2 males and 2 females; Supplementary Fig. 3e: control = 2 males and 2 females, Brn1/2-cKO = 3 males and 1 female; Supplementary Fig. 3f: control = 2 males and 1 female, Brn1/2-cKO = 3 males; Supplementary Fig. 3i: control = 3 males and 1 female, Brn1/2-cKO = 2 males and 1 female; Supplementary Fig. 4b-g: E12.5: control = 2 males and 1 female, Brn1/2-cKO = 1 male and 2 females; E14.5: control = 2 males and 2 females, Brn1/2-cKO = 3 males and 2 females; Supplementary Fig. 4i-n: E12.5: control = 2 males and 2 females, Brn1/2-cKO = 2 males and 1 female; E14.5: control = 2 males and 1 female, Brn1/2-cKO = 3 males and 1 female.
Field-collected samples	This study did not involve samples collected from the field.
Ethics oversight	All animal experiments adhered to the guidelines of the National Institute of Health and were approved by the Institutional Animal Care and Use Committee at Johns Hopkins University School of Medicine.

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

Plants

Seed stocks	*Report on the source of all seed stocks or other plant material used. If applicable, state the seed stock centre and catalogue number. If plant specimens were collected from the field, describe the collection location, date and sampling procedures.*
Novel plant genotypes	*Describe the methods by which all novel plant genotypes were produced. This includes those generated by transgenic approaches, gene editing, chemical/radiation-based mutagenesis and hybridization. For transgenic lines, describe the transformation method, the number of independent lines analyzed and the generation upon which experiments were performed. For gene-edited lines, describe the editor used, the endogenous sequence targeted for editing, the targeting guide RNA sequence (if applicable) and how the editor was applied.*
Authentication	*Describe any authentication procedures for each seed stock used or novel genotype generated. Describe any experiments used to assess the effect of a mutation and, where applicable, how potential secondary effects (e.g. second site T-DNA insertions, mosiacism, off-target gene editing) were examined.*