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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 Microscopy Software(ZEN 2.6) were used for all the immunofluorescence image acquisition; Zyla 4.2+ sCMOS were used for GCaMP fluorescence signal acquisition of widefiled imaging; Leica Application Suite X (LAS X 3.7.4 23463) were used for all the wild field brain image acquisition; QuantStudio™ 3 software (v1.3.0)were used for collecting all the RT-qPCR data; Luminescence Microplate Reader System were used for luciferase assay result collection and analysis;
Data analysis	All the images are analyzed by ImageJ (Java 1.8.0_322) and Qupath(v0.4.2 and v0.5.1); Binned analysis and the ggplots of bulkRNA-seq are generated by R-4.3.1; Venn diagrams generated by BioVenn(2007-2025 Tim Hulsen); GO analysis are performed by DAVID 2021 (Dec. 2021); scRNA-seq data count matrices were further processed using the Seurat R package version 4.0.5are processed by 10x Genomics cell ranger6.1.2 and cell cycle analyzed by scran R package1.22.1; Raw quantification data were initially processed by Excel (Microsoft,version 2501) and then followed analyzed and graphed with Prism (GraphPad Prism8).

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

Bulk RNA-seq datasets generated in this study are available under Gene Expression Omnibus accession number GSE287748; scRNA-seq datasets generated in this study are available under Gene Expression Omnibus accession number GSE288849.

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	No predetermination of sample sizes was carried out because our research is an exploratory study.
Data exclusions	No data were excluded during the process of data analyses.
Replication	All attempts at replication were successful. Mouse experiment was performed from at least 2 independent litters with at least 3 animals for each condition, and the sample sizes, biological and experimental duplications are listed in the figure legends. Images shown in all figures are representative of at least 3 independent brains. Human and chimpanzee cell experiments were performed from at least 2 independent differentiations, and the sample sizes, biological and duplications are listed in the figure legends. Images shown in all figures are representative of at least 3 independent experiments. Statistical analyses were included in figures for NPC islands or organoids, as standard for iPSC and organoid experiments. The data source tables also report statistics when each differentiation are included as the n value. Color coding was used for transparency of replication.To reduce technical variation, littermate samples across genotypes were processed in parallel for immunostaining with the same batches of reagents and identical imaging conditions.
Randomization	Allocation into groups was random. Data were assessed either by genotype for mouse or cell line, or experimental condition. To reflect biological variation, for the same experiment, we always collect samples at the same stage and include littermates as controls for each batch. All the adult animals are females for neural connectivity experiments, to avoid the variability between sex of animals.
Blinding	All analyses were performed by independent investigator blindly during data collection and data analysis by 1 or more investigators.

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) For immunofluorescence
Primary antibody: anti-NeuN (Cell Signaling,24307S,1:300 dilution); anti-Sox2 (Thermo,14-9811-82,1:1000 dilution); anti-Brn2 (Novus,NBP2-21585,1:400 dilution); anti-RORβ (R&D Systems, N7927,1:150 dilution); anti-Ctip2 (Abcam,AB18465,1:500 dilution); anti-Tbr1 (Abcam, AB31940,1:500 dilution); anti-Synapsin1 (Synaptic Systems, 106008, 1:200 dilution); DRAQ5 (Cell Signaling Technology, 4084S, 1:1000 dilution); anti-Sox9 (Millipore, AB5535,1:2000 dilution); anti-Olig2(Millipore; MABN50,1:500 dilution); anti-NeuroD2 (Abcam, #AB104430, 1:300 dilution); anti-PH3 (Millipore, 06-570,1:1000 dilution); anti-Tbr2 (Abcam,ab183991,1:1000 dilution); anti-Tuj1 (Covance,MM5-435P,1:1500 dilution); anti-BrdU (Abcam,ab6326,1:200 dilution); anti-BrdU (Cell Signaling,5292S, 1:300 dilution); anti-Ki67 (Cell signaling,12202,1:300 dilution); anti-Ki67 (Cell signaling,9449S,1:300 dilution); anti-RFP (Rockland,600-401-379,1:2000 dilution); anti-GFP (Abcam,ab13970,1:1000 dilution; anti-OCT4 (Abcam,ab19857,1:300 dilution); DAPI(Thermo Fisher,D1306,1:1000).
Secondary antibody:
Alexa Fluor 594 goat anti-rabbit (Thermo Fisher Scientific, A11037, 1:500 dilution); Alexa Fluor 594 goat anti-mouse(Thermo Fisher Scientific,A-11032,1:500 dilution); Alexa fluor 555 goat anti-rabbit (Thermo Fisher Scientific, A31572, 1:500 dilution); Alexa fluor 488 donkey anti-rat (Thermo Fisher Scientific,A21208, 1:500 dilution); Alexa fluor 488 goat anti-chicken (Thermo Fisher Scientific, SA5-10070,1:500 dilution);
Alexa fluor 488 goat anti-mouse (Thermo Fisher Scientific, A-11001,1:500 dilution); Alexa fluor 647 donkey anti-Rabbit(Thermo Fisher Scientific,A31573,1:500 dilution); Alexa fluor 647 donkey anti-mouse 647(Thermo Fisher Scientific,A31571 ,1:500 dilution).

2) For ChIP-qPCR
anti-H3 (Abcam, ab1791–2ug, 1:50 dilution), anti-H3K27me3 (Abcam, ab6002-3.5ug, 1:28 dilution) .

Validation

All the antibodies were validated by other published studies.
1) For immunofluorescence
Primary antibody: NeuN (Cell Signaling,24307S, marker for post-mitotic neurons, 263 Citations); Sox2 (Thermo,14-9811-82, marker for stem cells, used to label Neuroepithelium cells and Radial Gila cells in this study,75 Citations); Brn2 (Novus,NBP2-21585,marker for layer2/3 excitatory neurons ,2 Citations); RORβ (R&D Systems, N7927, marker for layer4 excitatory neurons,4 citations); Ctip2 (Abcam,AB18465, marker for layer5 excitatory neurons,840 Citations); Tbr1 (Abcam, AB31940, marker for layer6 excitatory neurons,475 Citations); Synapsin1 (Synaptic Systems, markers for synapse, 2 citation); DRAQ5 (Cell Signaling Technology, 4084S, staining of the nucleus, 8 citations); Sox9 (Millipore, AB5535,marker for astrocytes, PMID: 34171291); Olig2(Millipore; MABN50,marker for oligodendrocytes, 272 citations); NeuroD2 (Abcam, #AB104430, marker for newborn cortical pyramidal neurons,26 Citations); PH3 (Millipore, 06-570,marker for mitosis, 2649 Citations); Tbr2 (Abcam,ab183991,marker for intermediate progenitors, 81 Citations); Tuj1 (Covance,MM5-435P,marker for neurons, 473 Citations); BrdU (Abcam,ab6326, synthetic chemical to study cell proliferation in living tissues,1920 citations); BrdU (Cell Signaling,5292S, synthetic chemical used to study cell proliferation in living tissues, 221 citations); Ki67 (Cell signaling,12202,marker for dividing cells, 742 Citations); Ki67 (Cell signaling,9449S, marker for dividing cells,760 citations); RFP (Rockland,600-401-379, antibody to detect Red Fluorescent Protein,1187 citations); GFP (Abcam,ab13970, antibody to detect Green Fluorescent Protein,40 citations); OCT4 (Abcam,ab19857,marker for pluripotent stem cells ,560 citations). DAPI (Thermo Fisher,D1306, staining of the nucleus,2303 citations).
Secondary antibody:
Alexa Fluor 594 goat anti-rabbit (Thermo Fisher Scientific, A11037,2429 citations); Alexa Fluor 594 goat anti-mouse(Thermo Fisher Scientific,A-11032
2109 citations); Alexa fluor 555 goat anti-rabbit (Thermo Fisher Scientific, A31572, 2430 citations); Alexa fluor 488 donkey anti-rat (Thermo Fisher Scientific,A21208,1654 citations); Alexa fluor 488 goat anti-chicken (Thermo Fisher Scientific, SA5-10070,22 citations); Alexa fluor 488 goat anti-mouse (Thermo Fisher Scientific, A-11001,9007 citations); Alexa fluor 647 donkey anti-Rabbit(Thermo Fisher Scientific,A31573,2838 citations); Alexa fluor 647 donkey anti-mouse 647(Thermo Fisher Scientific,A31571,2250 citations)

2) For ChIP-qPCR
anti-H3 (Abcam, ab1791, ChIP grade rabbit polyclonal antibody used in Histone H3 western blotting, suitable for human, mouse and rat samples.4737 citations), anti-H3K27me3 (Abcam, ab6002, ChIP grade mouse monoclonal antibody that is used in Histone H3 (Tri Methyl-K27) western blotting for human and mouse samples. 981 citations).

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	ESC-WA09(human), RRID:CVCL_U176, female;
	iPSC-C3649(chimpanzee), RRID:CVCL_1G31,male;
	iPSC-8799(human),female;
	HEK293T,RRID:CVCL_0063, female;
	Neuroblastoma-SH SY5Y(human),RRID:CVCL_0019,female.

Authentication	iPSCs were authenticated by the original owner and WA09 were directly purchased from ATTC where they were authenticated. All the original human WA09 cells, iPSCs and edited cell lines were authenticated by OCT4 staining / morphology and Karyotyping. All the chimpanzee iPS cells and edited iPS cell lines were authenticated by OCT4 staining and morphlogy. HEK293T and SH SY5Y were authenticated by morphology.
Mycoplasma contamination	All cell lines used in this study tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	not included in this study.

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	Transgenic mice(HARE5-hs/hs; HARE5-cKO;Emx-1 Cre; HARE5::EGFP-PEST;TCF/LEF:H2B-EGFPTg/+; Tbr2-EGFPTg/+; Tbr2-Flp; Ai65-Frt) are C57BL/6J background; mFzd8 overexpression experiment is on CD1 background. Samples ages collected: E10.5, E12.5, E14.5, E15.5, P0, P21,P30, 2-4 months adults.
Wild animals	This study did not involve wild animals
Reporting on sex	For the neural connectivity experiments, to exclude the differences due to sex, all the adult animals were females; Other experiments were selected randomly on sex.
Field-collected samples	This study did not involve samples collected from field.
Ethics oversight	All mice experiments were approved by Duke IACUC and followed the guidelines from the Division of Laboratory Animal Resources from Duke 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	N/A
Novel plant genotypes	N/A
Authentication	N/A