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Reporting Summary

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10.24433/CO.7867348.v1).

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For	ali statisticai ani	alyses, 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 stateme	nt 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.			
\boxtimes	A descripti	ion of all covariates tested		
\boxtimes	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>			
\boxtimes	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings			
\boxtimes	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes			
\boxtimes	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 <u>statistics for biologists</u> contains articles on many of the points above.				
Software and code				
Poli	cy information a	about <u>availability of computer code</u>		
Da	ata collection	Synapse (Tucker-Davis Technologies, Inc.) VersaStudio		
Da	ata analysis	Synapse (Tucker-Davis Technologies, Inc.)		

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.

Other analyses were done with custom Matlab code. The source code is available under the license of CCO at Code Ocean (https://doi.org/

Data

Blinding

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 main data supporting the results in this study are available within the paper and its Supplementary Information. The datasets generated during the study are too large to be publicly shared. Source data are available at Figshare (https://doi.org/10.6084/m9.figshare.24764508). Source data are provided with this paper for reproducing all Figures in the manuscript and Supplementary Information.

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. The study did not involve human research participants. Reporting on sex and gender Reporting on race, ethnicity, or The study did not involve human research participants. other socially relevant groupings Population characteristics The study did not involve human research participants. Recruitment The study did not involve human research participants. The study did not involve human research participants. Ethics oversight 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. X Life sciences Behavioural & social sciences Ecological, evolutionary & environmental sciences For a reference copy of the document with all sections, see <u>nature.com/documents/nr-reporting-summary-flat.pdf</u> Life sciences study design All studies must disclose on these points even when the disclosure is negative. Sample sizes were determined by the number of biological and technical replicates necessary to observe the effect was real. Sample size Data exclusions There were no exclusions. At least three organoids were performed in each case of this study, and all experimental findings were reliably reproduced at least three Replication All samples were allocated randomly into experimental groups. Randomization

Reporting for specific materials, systems and methods

Blinding was not relevant, because a blinding process wouldn't influence the sampling result.

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 & experi	mental systems Methods
Animals and oth Clinical data	ines ChIP-seq Flow cytometry and archaeology MRI-based neuroimaging
Dual use resear Plants	ch of concern
Antibodies	
Antibodies used	Antibody name (supplier; catalog number): Mouse Anti-TUJ1 (BioLegend; 801202), Rabbit Anti-TUJ1 (Cell Signaling Technology; 5568S), Rabbit Anti-SOX2 (Millipore; AB5603), Rabbit Anti-MAP2 (Cell Signaling Technology; 4542S), Mouse Anti-GFAP (Millipore; MAB3402), Rabbit Anti-VGLUT1 (Synaptic Systems; 135 302), Rat Anti-Ctip2 (Abcam; ab18465), Mouse Anti-Human SATB2 (Abcam; ab51502), Mouse Anti-Nestin (Millipore; MAB5326), Mouse Anti-Phospho-Tau (Thermo Fisher Scientific; MN1020), Alexa Fluor 488 goat anti-mouse IgG (Thermo Fisher; A11001), Alexa Fluor 488 goat anti-rabbit IgG (Thermo Fisher; A11008), Alexa Fluor 488 goat anti-rabbit IgG (Thermo Fisher; A11005), Alexa Fluor 594 goat anti-mouse IgG (Thermo Fisher; A11012), Alexa Flour 555 goat anti-mouse IgG1 (Thermo Fisher; A21127), Alexa Flour 488 goat anti-Mouse IgG2a (Thermo Fisher; A21131)
Validation	All antibodies listed above are commercially available and have been verified by many references provided on the website of the companies that sell antibodies (links below).
	Mouse Anti-TUJ1 (BioLegend; 801202) Validation Refs. from the manufacturer's datasheet: https://www.biolegend.com/fr-lu/explore-new-products/purified-anti-tubulin-beta-3-tubb3-antibody-11580?pdf=true&displayInline=true&leftRightMargin=15&topBottomMargin=15&filename=Purified%20anti-Tubulin%20%CE%B2%203%20(TUBB3)%20Antibody.pdf&v=20240814063131
	Rabbit Anti-TUJ1 (Cell Signaling Technology; 5568S) Validation Refs. from the manufacturer's datasheet: https://www.cellsignal.com/products/5568/datasheet? images=1&srsltid=AfmBOorgFXlgrO0yWfNKZNOYn2Qr8dxEki77brb9D0mPJrspXsOT5H0P
	Rabbit Anti-SOX2 (Millipore; AB5603) Validation Refs. from the manufacturer's datasheet: https://www.sigmaaldrich.com/KR/ko/product/mm/ab5603? srsltid=AfmBOorKAaotQpgxi9b6iv7x_WbbqJNCcxVYkI7lmOAeQCJs-ZaLG1WJ
	Rabbit Anti-MAP2 (Cell Signaling Technology; 4542S) Validation Refs. from the manufacturer's datasheet: https://www.cellsignal.com/datasheet.jsp? productId=4542&images=1&srsItid=AfmBOookdlcV-B_ycSr_JCCfZMZN77MLjrBwkPnk7kgR-zFnEqRqcY0r
	Mouse Anti-GFAP (Millipore; MAB3402) Validation Refs. from the manufacturer's datasheet: https://www.merckmillipore.com/KR/ko/product/Anti-Glial-Fibrillary-Acidic-Protein-Antibody-clone-GA5,MM_NF-MAB3402
	Rabbit Anti-VGLUT1 (Synaptic Systems; 135 302) Validation Refs. from the manufacturer's datasheet: https://sysy.com/product/135302
	Rat Anti-Ctip2 (Abcam; ab18465) Validation Refs. from the manufacturer's datasheet: https://www.abcam.com/en-us/products/primary-antibodies/ctip2-antibody-25b6-ab18465#tab=datasheet
	Mouse Anti-Human SATB2 (Abcam; ab51502) Validation Refs. from the manufacturer's datasheet: https://www.abcam.com/en-us/products/primary-antibodies/satb1-satb2-antibody-satba4b10-c-terminal-ab51502
	Mouse Anti-Nestin (Millipore; MAB5326) Validation Refs. from the manufacturer's datasheet: https://www.merckmillipore.com/KR/ko/product/Anti-Nestin-Antibody-clone-10C2,MM_NF-MAB5326
	Mouse Anti-Phospho-Tau (Thermo Fisher Scientific; MN1020) Validation Refs. from the manufacturer's datasheet: https://www.thermofisher.com/antibody/product/Phospho-Tau-Ser202-Thr205-Antibody-clone-AT8-Monoclonal/MN1020
	Alexa Fluor 488 goat anti-mouse IgG (Thermo Fisher; A11001) Validation Refs. from the manufacturer's datasheet; https://www.thermofisher.com/order/genome-database/datasheetPdf?

 $product type = antibody \& product subtype = antibody_secondary \& product Id = A-11001 \& version = Local \\$

Alexa Fluor 488 goat anti-rabbit IgG (ThermoFisher; A11008)

Validation Refs. from the manufacturer's datasheet: https://www.thermofisher.com/order/genome-database/dataSheetPdf? $product type=antibody \& product subtype=antibody_secondary \& product Id=A-11008 \& version=Local type=antibody & product Subtype=antibody & product Subtype$

Alexa Fluor 488 goat anti-rat IgG (Thermo Fisher; A11006)

Validation Refs. from the manufacturer's datasheet: https://www.thermofisher.com/order/genome-database/dataSheetPdf? $product type=antibody \& product subtype=antibody_secondary \& product Id=A-11006 \& version=Local type=antibody & product Subtype=antibody & product Subtype$

Alexa Fluor 594 goat anti-mouse IgG (Thermo Fisher; A11005)

Validation Refs. from the manufacturer's datasheet:

https://www.thermofisher.com/antibody/product/Goat-anti-Mouse-IgG-H-L-Cross-Adsorbed-Secondary-Antibody-Polyclonal/secondary-Antibody-Polyclonal-secondary-secondary-secondary-secondary-secondary-secondary-secondaryA-11005

Alexa Fluor 594 goat anti-rabbit IgG (Thermo Fisher; A11012)

Validation Refs. from the manufacturer's datasheet:

https://www.thermofisher.com/antibody/product/Goat-anti-Rabbit-lgG-H-L-Cross-Adsorbed-Secondary-Antibody-Polyclonal/antibodyA-11012

Alexa Flour 555 goat anti-mouse IgG1 (Thermo Fisher; A21127)

Validation Refs. from the manufacturer's datasheet:

https://www.thermofisher.com/antibody/product/Goat-anti-Mouse-IgG1-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21127

Alexa Flour 488 goat anti-Mouse IgG2a (Thermo Fisher; A21131)

Validation Refs. from the manufacturer's datasheet:

https://www.thermofisher.com/antibody/product/Goat-anti-Mouse-IgG2a-Cross-Adsorbed-Secondary-Antibody-Polyclonal/A-21131

Eukaryotic cell lines

Cell line source(s)

Policy information about cell lines and Sex and Gender in Research

The human induced pluripotent stem cells (hiPSCs; Female; KYOU-DXR0109B) were acquired from American Type Culture Collection.

Authentication The use of human iPSCs was approved by the Institutional Review Board (IRB) of Yonsei University (Permit Number:

7001988-202309-BR-1066-02E).

The hiPSCs were regularly checked for mycoplasma contamination. Mycoplasma contamination

Commonly misidentified lines (See ICLAC register)

No commonly misidentified lines were used.

Plants

Seed stocks The study did not involve plant samples.

Novel plant genotypes

The study did not involve plant samples.

Authentication

The study did not involve plant samples.