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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	Cerebral blood flow by laser Doppler fluometer and blood pressure: PowerLab/LabChart 8 (ADINSTRUMENTS), Cerebral blood flow by laser speckle imaging: Omegazone imaging Software (Omegawave), Cytoplasmic Ca2+: IPLab Imaging Software (BD Biosciences), Multi-photon imaging: Olympus Fluoview; MATLAB code; ImageJ, Image processing: ImageJ (Version 1.52a), Behavioral testes: AnyMaze (Stoelting)
Data analysis	Histological and biochemical analyses were performed in a blinded fashion. Data are expressed as means±SEM. Data and Image analysis were done with ImageJ (NIH), MATLAB (R2018a, MathWorks, Inc.), or Prism (version 8.1.0, GraphPad Software, Inc.). Two-group comparisons were analyzed by the two-tailed t-test. Multiple comparisons were evaluated by the analysis of variance and Tukey's test. Differences were considered statistically significant for probability values less than 0.05.

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

All the data that support the findings of the study are presented in the figures of manuscript and are available upon request from the corresponding authors. There are no restrictions on data availability.

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	The sample size for the in vivo CBF, behavior, or MRI experiments was determined with power analysis using G*Power program (Version 3.1.9.2.) with the following parameters: t-tests: differences between two independent means (two groups), two tailed, effect size d: 2.36 or 3.56 (based on preliminary data and previous experiences), alpha error prob: 0.05, Power: 0.80 leading to a total sample size of 5-10 animals per group. Ex vivo experiments (immunoprecipitation, Ca2+ imaging, NO production, and immunofluorescent quantification) were performed with N=4-10 mice per group based on previous similar experiences and preliminary experiments with power of 80-90%.
Data exclusions	No data were excluded.
Replication	All in vivo experiments were performed on more than 5 independent biological replicates. All ex vivo or in vitro experiments were replicated in >4 independent animals or samples per experiments. All attempts at replication were successful.
Randomization	In vivo and ex vivo experiments were performed in randomly assigned mice or samples using Random number generator at www.random.org.
Blinding	Blinding was not applied to studies involving plasmid constructs and transient transfections and co-immunoprecipitation in HEK293T cells because plasmids were constructed knowingly and carefully and cell lines needed to be transfected and maintained separately. However, data analysis were performed blind whenever possible.

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

Antibodies

Antibodies used

<Primary antibodies for immunofluorescent staining>
Name of antibody/Clone/Host/Antibody type/Catalog number/IgG subclass/Working dilution/Suppliers
AQP4/D1F8E/Rabbit/Monoclonal/59678/IgG/1:200/Cell Signaling
CD31/MEC 13.3/Rat/Monoclonal/553370/IgG2a, k/1:30/BD Biosciences
GFAP/G-A-5/Mouse/Monoclonal/G3893/IgG1/1:1000/Sigma
Iba-1/---/Rabbit/Policlonal/019-19741/---/1:500/Wako/Chemical
MBP/12/Rat/Monoclonal/MAB386/IgG2a/1:500/EMD Millipore
MAP2/---/Rabbit/Policlonal/M3696/---/1:500/Sigma
NeuN/EPR12763/Rabbit/Monoclonal/ab177487/IgG/1:100/abcam
Phospho-Tau/AT8/Mouse/Monoclonal/MN1020/IgG1/1:200/ThermoFisher

<Primary antibodies for Western Blotting>
CamKII alpha/Cba-2/Mouse/Monoclonal/13-7300/IgG2a, k/1:500/ThermoFisher
GAPDH/6C5/Mouse/Monoclonal/CB1001/IgG1/1:500/EMD Millipore
GluN2B/13/Mouse/Monoclonal/610416/IgG2b/1:500/BD Biosciences
MEK1/2/---/Rabbit/Polyclonal/9122/---/1:1000/Cell Signaling
nNOS/---/Rabbit/Polyclonal/BML-SA227/---/1:2000/Enzo Life Sciences
PSD95/16/Mouse/Monoclonal/610496/IgG1/1:250/BD Biosciences
Phospho-Tau/AT8/Mouse/Monoclonal/MN1020/IgG1/1:500/ThermoFisher

Phospho-Tau/PHF-13/Mouse/Monoclonal/9632/IgG2b/1:1000/Cell Signaling
Total Tau/Tau46/Mouse/Monoclonal/4019/IgG1/1:1000/Cell Signaling
Total Tau/TAU-5/Mouse/Monoclonal/AHB0042/IgG1/1:200/ThermoFisher
Synaptophysin/D35E4/Rabbit/Monoclonal/5461S/IgG/1:1000/Cell Signaling

<Primary antibodies for immunoprecipitation>
nNOS/---/Rabbit/Polyclonal/PA3-032A/---/4 µg/ThermoFisher
PSD95/6G6-1C9/Mouse/Monoclonal/MA1-045/IgG2a/4 µg/ThermoFisher
Total Tau/TAU-5/Mouse/Monoclonal/AHB0042/IgG1/4 µg/ThermoFisher
Isotype control/RMG2a-62/Rat/Monoclonal/407102/IgG2a/4 µg/BioLegend
Isotype control/RMG1-1/Rat/Monoclonal/406602/IgG1/4 µg/BioLegend
Isotype control/Poly29108/Rabbit/Polyclonal/910801/---/4 µg/BioLegend

Validation

According to the specification sheets provided by manufacturers and extensive available literature, as well as our own studies, all antibodies used in this study for immunofluorescent stainings, Western Blotting, and immunoprecipitation have been tested for specific reactivity against respective antigens in the target species, by either using a) tagged over-expression controls, b) comparison with knockdown or knockout controls, c) blocking antibody controls, d) omission of the primary and/or secondary antibody, or e) fractionation controls.

<Primary antibodies for immunofluorescent staining>
- AQP4 was validated by the manufacturer (Cell Signaling) using various methods, including Western blot analysis to demonstrate specific bands of the appropriate molecular weight(s) with minimal cross-reacting bands, the use of blocking peptides to verify specificity and rule out Fc-mediated binding and other non-specific staining, thorough lot testing to ensure the reproducibility necessary for accurate immunohistochemistry results, predetermined dilutions, and specified protocols and control reagents. The concentration used in the current study was determined by using a series of dilution in a titration experiment.
- CD31 was validated by the manufacturer (BD Biosciences) and extensively verified in the literatures, including our own studies (PMID: 7956830; PMID: 30232327; PMID: 8082649). The concentration used in the current study was determined in our previous study (PMID: 30232327).
- GFAP was validated by the manufacturer (Sigma). The antibody is suitable for immunohistochemical staining. The isotype is determined using ImmunoTypeTM Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2). The concentration used in the current study was determined in our previous study (PMID: 28515043).
- Iba-1 was validated by the manufacturer (Wako Chemical) and is well verified in the literatures for immunofluorescent staining, including our own studies (PMID: 28802038; PMID: 27135602; PMID: 30232327; PMID: 28515043). The concentration used in the current study was determined in our previous studies (PMID: 30232327; PMID: 28515043).
- MBP was verified by the manufacturer (EMD Millipore). The antibody reacts with MBP from all species tested. It reacts weakly with peptides ending in the phe 91 where the 91-92 phe-phe bond is broken. Synthetic peptide 82-99 reacts very well as does intact MBP. Mapped to the region DENPVV. The concentration used in the current study was determined in our previous study (PMID: 30232327).
-MAP2 was validated by manufacturer (Sigma). For specificity, Clone 3C19 is a ZooMAb rabbit recombinant monoclonal antibody that specifically detects human Microtubule-associated protein 2. It targets an epitope with in 17 amino acids from the C-terminal half. The concentration used in the current study was determined by using a series of dilution in a titration experiment.
- NeuN was validated by the manufacturer (abcam). The exact sequence is proprietary. The concentration used in the current study was determined based on our previous study (PMID: 17611261).
- Phospho-Tau (AT8) was validated by the manufacturer (ThermoFisher). AT8 immunogen is partially purified human PHF-Tau and detects PHF-tau (Ser202/Thr205)a which has a predicted molecular weight of approximately 79 kDa. The concentration used in the current study was determined by using a series of dilution in a titration experiment.

<Primary antibodies for Western Blotting>
- CaMKII alpha (Thermo Fisher): Antibody specificity was demonstrated by the vendor by detection of differential basal expression of the target across tissues tested owing to their inherent genetic constitution. Expression of CAMKII alpha was observed in mouse brain, but not in mouse skeletal muscle and mouse heart, which are negative for the CaMKII alpha gene.
- GAPDH (EMD Millipore): Antibody was validated for use in Western Blot by the vendor to recognize mouse GAPDH. In addition, our study shows specificity for the cytosolic protein GAPDH by detecting a band only in cytosolic fractions, and not in membrane or PSD (Extended Supplementary Figure 10D).
- GluN2B (BD Biosciences): Verified by the vendor by western blot analysis to detect mouse GluN2B. In addition, as shown in our study, a band with GluN2B, as a PSD-associated protein, is specifically detected in PSD fractions, and not in cytosol or membrane (Supplementary Figure 10B).
- MEK1/2 (Cell Signaling): Antibody was validated for use in Western Blot of mouse samples by the vendor. In addition, as shown in our study, a band with MEK1/2, as a cytosolic protein, is only detected in cytosolic fractions, and not in membrane or PSD (Supplementary Figure 10B).
- nNOS (Enzo Life Sciences): Western blot analysis by the vendor with nNOS antibody using whole cell extracts from mouse brain showed a prominent band that was abolished with pre-incubation with a blocking control peptide.
- PSD95 (BD Biosciences): Antibody was validated for use in Western Blot of mouse samples by the vendor. In addition, our study shows specificity for the PSD protein PSD95 by detecting a band only in PSD fractions, and not in cytosol or membrane (Extended - Supplementary Figure 10D). The antibody also specifically detects the over-expressed protein (Figures 8B and D).
- Phospho-Tau/AT8 (Thermo Fisher): Extensively verified in the literature to detect human Tau phosphorylated at Ser 202/205. In addition, the AT8 Tau antibody was validated in our study for phospho-specificity by treating samples with Lambda phosphatase (Figure 8A).
- Phospho-Tau/PHF-13 (Cell Signaling): Verified by the vendor by western blot analysis of extracts from mouse brain. The phospho-specificity of the antibody was verified by peptide blocking using no peptide, phospho-peptide or nonphospho-peptide.
- Total Tau46 (Cell Signaling): Verified by the vendor for specificity by Western blot analysis of Tau WT and Tau KO (-/-) mouse brain.
- Total Tau-5 (ThermoFisher): Antibody specificity was demonstrated by the vendor by detection of differential basal expression of the target across mouse brain, mouse kidney, and mouse lung owing to their inherent genetic constitution. Relative

expression of Tau was observed in mouse brain as compared to kidney and lung tissues in Western Blot. The antibody also detects human Tau, as shown in our study by overexpression experiments (Figure 8C).
- Synaptophysin (Cell Signaling): Was shown by vendor to detect mouse synaptophysin using Western blot analysis of extracts from mouse brain. In addition, as shown in our study, as a pre-synaptic membrane-specific protein, a band with synaptophysin antibody is only detected in presynaptic membrane fractions, but not in the PSD (Extended Supplementary Figure 10D).

<Primary antibodies for immunoprecipitation>
All used antibodies for immunoprecipitation were validated by vendors for species and antigen reactivity, as well as in each immunoprecipitation experiment in our study for specificity with IgG control antibodies.
- nNOS (Thermo Fisher): Western blot analysis against mouse brain tissue lysates by the vendor verified reactivity for mouse nNOS. In addition, in our study this antibody's specificity has been verified in immunoprecipitation experiments with an IgG control antibody (Figure 7F).
- PSD95 (Thermo Fisher): Antibody specificity was demonstrated by the vendor by detection of differential basal expression of the target across tissues owing to their inherent genetic constitution. Relative expression of PSD-95 was observed in mouse brain in comparison to mouse skeletal muscle and mouse liver, which are devoid of PSD-95. In addition, in our study this antibody's specificity has been verified in immunoprecipitation experiments with an IgG control antibody (Figures 7E, 8B, 8D).
- Total Tau-5, (Thermo Fisher): Antibody specificity was demonstrated by the vendor by detection of differential basal expression of the target across mouse brain, mouse kidney, and mouse lung owing to their inherent genetic constitution. Relative expression of Tau was observed in mouse brain as compared to kidney and lung tissues in Western Blot. The antibody also detects human Tau, as shown in our study by overexpression experiments (Figure 8C). In addition, in our study this antibody's specificity has been verified in immuno-precipitation experiments with an IgG control antibody (Figure 8C).

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Human embryonic kidney (HEK) 293T cells from American Tissue Culture Collection (ATCC; CRL-11268)
Authentication	General information and authentication procedures for the HEK 293T/17 cell lines are available at the ATCC websites: https://www.atcc.org/products/all/CRL-11268.aspx#generalinformation
Mycoplasma contamination	HEK 293T cell line was tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	None used

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	All procedures were performed according to the ARRIVE guidelines. Experiments were performed in two tau transgenic mouse lines: rTg4510 (P301L) and PS19 (P301S) (Jackson Lab stock# 008169) mice. The bi-transgenic rTg4510 mice were generated with conditional tau P301L expression driven by the CaMKIIalpha promoter by crossing responder (The Jackson Lab stock# 015815) and activator (Jackson Lab stock# 007004) transgenic lines. All the experiments were performed at the age of 2-8 months. All tau transgenic mice and wild type littermates were males.
Wild animals	Wilds animals were not investigated in the study.
Field-collected samples	Field-collected samples were not studied in the study.
Ethics oversight	All procedures were approved by the Institutional Animal Care and Use Committee of Weill Cornell Medicine.

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

Magnetic resonance imaging

Experimental design

Design type	Anatomical and perfusion MRI
Design specifications	Anesthesia with isoflurane. Total scan time per animal (ASL+anatomical image): About 40 min
Behavioral performance measures	Behavioral performance was not measured.

Acquisition

Imaging type(s)	ASL (perfusion imaging); T2-weighted (anatomical imaging)
Field strength	7.0 Tesla 70/30 Bruker Biospec small animal MRI system
Sequence & imaging parameters	ASL: Three averages of one axial slice were acquired with a field of view of 15 × 15 mm, spatial resolution of 0.234 × 0.234 × 2 mm3, echo time TE of 5.368 ms, effective TE of 26.84 ms, repeat time TR of 10 s, and a RARE factor of 36. For

computation of CBF (ml/100g/min), the Bruker ASL perfusion processing macro was used. Turbo-RARE anatomical images were acquired with the same field-of-view and orientation as the ASL images (resolution = 0.078 × 0.078 × 1 mm3, effective TE = 48 ms, TR = 2200 ms, and a RARE factor of 10).

Area of acquisition

Anatomical localizer images were acquired to find the transversal slice corresponding to the somatosensory cortex (from -1.22 to -1.70 mm from Bregma). This position was used for subsequent ASL imaging, which was based on a FAIR-RARE pulse sequence labeling the inflowing blood by global inversion of the equilibrium magnetization.

Diffusion MRI ☐ Used ☒ Not used

Preprocessing

Preprocessing software: Bruker ASL perfusion processing macro; ImageJ

Normalization: N/A

Normalization template: N/A

Noise and artifact removal: Thresholding to remove regions not belonging to brain parenchyma

Volume censoring: N/A

Statistical modeling & inference

Model type and settings: N/A

Effect(s) tested: N/A

Specify type of analysis: ☐ Whole brain ☒ ROI-based ☐ Both

Anatomical location(s): Somatosensory cortex (from -1.22 to -1.70 mm from Bregma)

Statistic type for inference (See Eklund et al. 2016): Data were analyzed by one-way or two-way ANOVA plus Tukey's test for multiple comparisons.

Correction: N/A

Models & analysis

n/a	Involved in the study
☒	☐ Functional and/or effective connectivity
☒	☐ Graph analysis
☒	☐ Multivariate modeling or predictive analysis