

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 (<i>n</i>) 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 <i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i>
☒	☐ 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. <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>
☒	☐ 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 <i>d</i> , Pearson's <i>r</i>), 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	Tools and software for data collection have been described in: Zheng Z, Own CS, Wanner AA, et al. Fast imaging of millimeter-scale areas with beam deflection transmission electron microscopy. Nat Commun. 2024;15(1):6860. doi:10.1038/s41467-024-50846-4
Data analysis	Transforms and registrations of mouse template brains: https://github.com/navis-org/navis-mousebrains Reconstruction pipeline code: https://github.com/seung-lab/seuron Code for reading and writing of image volumes: https://github.com/seung-lab/cloud-volume The analysis code: https://github.com/seung-lab/ca3_paper Connectome Annotation Versioning Engine: https://github.com/CAVEconnectome

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 EM image (rendered at 18 nm/pixel), segmentation, and annotation data can be accessed at <https://pyr.ai>

A list of all identified cell types, along with links to the segmentation and EM data, can be found at https://codex.flywire.ai/research/mouse_ca3_explorer. An account is required, but access is open to all users.

Dataset tutorials:

<https://blog.pyr.ai/2025/01/08/cell-and-anatomy-guide-to-ca3/>

<https://blog.pyr.ai/2025/02/11/mossy-fiber-challenge/>

Proofreading tutorials:

<https://blog.pyr.ai/2024/12/20/proofreading-101-climb-into-spelunker/>

<https://blog.pyr.ai/2025/02/07/spelunker-102-advanced-proofreading-tips/>

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](https://www.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 sample-size calculation was performed a priori. Sample sizes (number of neurons or connections tested) match or exceed previous studies of the same brain region.

Data exclusions

Hippocampus tissues of 4 animals from both sexes were screened, and one sample with the best staining quality was used for sectioning and imaging.

Replication

Connectomic data were collected only from one animal due to time and cost constraints. However, all proofreading in the volume was used to increase power with regard to number of neurons or connections.

Randomization

No randomization is performed since our study did not include multiple predefined experimental groups for sample allocation.

Blinding

No blinding is performed during data collection since our study did not include predefined experimental groups for sample allocation. Manual proofreading of the data is blinded to the goals of the analysis. The analysis is performed unblinded, however, the same process is applied to all control and sample groups.

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

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	A 4-month-old female mouse (Mus musculus; C57BL/6J-Tg(Thy1-GCaMP6f)GP5.3Dkim/J; The Jackson Laboratory strain no. 028280) was housed under a reverse light cycle (lights off: 8:00 AM–8:00 PM; lights on: 8:00 PM–8:00 AM) at 70 ± 2 °F temperature and 50 ± 10% humidity.
Wild animals	The study did not involve wild animals.
Reporting on sex	One female mouse was used. Animals were randomly recruited to the study with respect to sex.
Field-collected samples	The study did not involve field-collected samples.
Ethics oversight	Institutional Animal Care and Use Committee at Princeton University

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