

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](#).

Statistical parameters

When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. 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
- ☐ ☒ An indication of 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 statistics 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
- ☐ ☒ Clearly defined error bars
State explicitly what error bars represent (e.g. SD, SE, CI)

Our web collection on [statistics for biologists](#) may be useful.

Software and code

Policy information about [availability of computer code](#)

Data collection

A custom software package, RTAOI (Real-Time All-Optical Interface), developed using VB.NET Framework 4.5, Visual studio 2013 (Microsoft), CUDA toolkit 8 (NVIDIA), NI-DAQmx 15.5 (National Instruments), together with PrairieLink (Bruker Corporation) and Prairie View 5.4 (Bruker Corporation) was used for microscope control and imaging data collection. Blink SDK (Meadowlark Optics) was used for spatial light modulator control. PackIO (Watson et al, bioRxiv, 2016) was used for read/write electrical signals from National Instruments devices.

Data analysis

The custom software package (RTAOI, as mentioned above) was used for online data analysis; MATLAB2016a and Python 3.5 were used for off-line data analysis; Suite2P (Pachitariu et al, bioRxiv, 2016) was used for off-line analysis of calcium imaging data.

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 upon request. 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

The original code, together with detailed instructions and demo data is available at the following Github link: <https://github.com/alloptical/ClosedLoop>; Correspondence and requests for raw data should be addressed to M.H. (m.hausser@ucl.ac.uk).

Field-specific reporting

Please select 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/authors/policies/ReportingSummary-flat.pdf](https://www.nature.com/authors/policies/ReportingSummary-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. In cases where statistics are reported, we used the minimum number of animals possible to obtain a reasonable estimate of the effect as per the "3R" guidelines required by law in the UK. See the first paragraph of the methods section.
Data exclusions	No samples were excluded except in obvious cases where experiments could no longer be performed due to poor viral expression or occluded chronic windows. In the activity clamp experiments, recordings were aborted and were not included in analysis when the baseline firing rate of the target cell went above 4 Hz. Trials in which the baseline spike rates and/or amplitudes decreased by more than 80%, and/or the spike widths (measured as the full width at half maximum) increased by more than 80% compared to those in the initial recording were not included in analysis (see Methods). Fig. 2i-l only includes experiments in which greater than 100 trigger-targets pairings were performed during conditioning on animals younger than 14 weeks old.
Replication	The reported statistics were based on sufficient sample size; number of replications are stated in the figure legends; experiments were done in batches rather than all at once. The range of the factors that could potentially limit the reproducibility of the experimental results (e.g. animal age, cell spontaneous rate) is provided (see Methods). Multiple ways of analysis were done on the same dataset to show that results were immune to analysis particularities (e.g. both inferred spike rate and calcium event rate were tested to show the change in the activity level in the conditioning experiments in Fig. 2m and Supplementary Fig.8); The exact details of the methods are provided in the methods section.
Randomization	No randomization was necessary since no experimental groups were formed.
Blinding	No blinding was necessary since no group allocation was performed.

Reporting for specific materials, systems and methods

Materials & experimental systems

n/a	Involved in the study
☒	☐ Unique biological materials
☒	☐ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology
☐	☒ Animals and other organisms
☒	☐ Human research participants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Animals and other organisms

Policy information about [studies involving animals](#); [ARRIVE guidelines](#) recommended for reporting animal research

Laboratory animals	Female wild-type adult mice, approximately 4 weeks to 8 weeks old were used in this study.
Wild animals	No wild animals were used.
Field-collected samples	No field-collected samples were used