nature portfolio

Corresponding author(s): Jianfeng Liu, Yan Zhang, Jean-Philippe Pin

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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	Cryo-EM data collection was performed using serialEM. IP1 measurements were acquired using PHERAstar FS (BMG Labtech, USA) with the program PHERAstar control Version 4.00 R4. and exported using MARS Data Analysis Software v3.01 R2 (BMG LABTECH, USA). Intracellular calcium release measurements were acquired using Flexstation III (Molecular Devices, Sunnyvale, CA, USA) with the SoftMax Pro Software v5.4.6.005 (Molecular Devices, Sunnyvale, CA, USA). BRET measurements were acquired using Mithras LB 940 multimode microplate reader (Berthold Technologies, Bad Wildbad, Germany) with the program MikroWin, Version 4.41; or PHERAstar FS with the program PHERAstar control Version 4.00 R4, or Infinite 200 PRO (Tecan, Switzerland) with the program i-control Version 1.12. Images of gel were collected using an Odyssey infrared scanner (LI-COR Biosciences, Lincoln, NE, USA).
Data analysis	The following software was used in this study: MotionCor2.1, gCTF, RELION 3.0-beta2, UCSF Chimera, UCSF ChimeraX, Coot, Phenix, Graphpad Prism 8, PAM (for smFRET data analysis).

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

All data generated during this study are included in this paper and its Supplementary Information. PDB and EMDB files have been created and will be available on publication. All the source data are provided. The data didn't include any clinical experiments.

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	Sample size for each experiments were determined based on standards for experimental cell biology, with a minimum of n = 3 biological independent replicates with sufficient reproducibility. All the presented data are mean ± SEM or representative results for at least three experiments. Information on the number of replicates and independent experiments that were performed for each measurement is provided in the manuscript.
Data exclusions	No data was systematically excluded.
Replication	Number of independent experiments and replicates are indicated in the legends to the figures. Experimental findings were reliably reproduced.
Randomization	No randomization was attempted or needed. Randomization was not necessary as the independent variables to be tested were sufficient for the functional interpretation within this study.
Blinding	Blinding was not applicable to this study. All experimental data were acquired using automated equipment and analyzed using computational softwares, eliminating human error and bias.

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

secondary anti-rabbit antibody(A6154, 1:1000, Sigma Aldrich).
Anti-His antibody (34660, 1:5000, QIAGEN) .
Monoclonal ANTI-FLAG® M2-Peroxidase antibody (A8592, 1:10000, Sigma Aldrich, Shanghai, China).
Monoclonal Anti-HA–Peroxidase antibody (3F10, 1:2000, Roche, Shanghai, China).

Validation

secondary anti-rabbit antibody Sigma Aldrich A6154, Validated by company (https://www.sigmaaldrich.cn/CN/zh/product/sigma/a6154)
Anti-His antibody QIAGEN 34660
Monoclonal ANTI-FLAG® M2-Peroxidase antibody Sigma Aldrich A8592, Validated by company (https://www.sigmaaldrich.cn/CN/zh/product/sigma/a8592)
Monoclonal Anti-HA–Peroxidase antibody Roche 3F10, Validated by company (https://www.sigmaaldrich.cn/CN/zh/product/roche/roahaha)

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)

HEK293 cells were obtained from Cell Resource Center of Shanghai Institute for Biological Sciences (Chinese Academy of Sciences, Shanghai, China). Hi5 cells were purchased from Expression Systems (Cat 94-001S).

Authentication

All of the cell lines are maintained by the supplier. No additional authentication was performed by the authors of this study.

Mycoplasma contamination

Cell lines are tested by manufacturer for contamination.

Commonly misidentified lines
(See ICLAC register)

No commonly misidentified cell lines were used.

Plants

Seed stocks

N/A

Novel plant genotypes

N/A

Authentication

N/A