nature portfolio

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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	Latitude S 1.3 - commercially available, SerialEM 3.8 - published and freely available
Data analysis	RELION 4.0 - published and freely available, UCSF pyem v0.5 - published and open source, CryoSPARC 4.0/4.1/4.4- commercially available, Coot 0.9- published and freely available, PHENIX 1.20 published and freely available, UCSF Chimera X 1.6 - published and freely available, Prism 10 -commercially available, PyMOL 2.4.0 - open source, APBS Tools 2.1 - open source, CHARMM-GUI - published and freely available, AMBER20 - commercially available, VMD 1.9.4 - published and freely available, MAFFT v7.475 - - published and open source.

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 atomic coordinates has been deposited in the Protein Data Bank (PDB) under accession codes 9DK9 (no ligand added-URAT1CS); 9DKA (BBR-URAT1CS); 9DKB (LESU-URAT1CS); and 9DKC (TD-3-URAT1CS), respectively. The cryo-EM maps has been deposited in the Electron Microscopy Data Bank (EMDB) under accession codes EMD-46948 (no ligand added-URAT1CS); EMD-46949 (BBR-URAT1CS); EMD-46950 (LESU-URAT1CS); and EMD-46951 (TD-3-URAT1CS), respectively.

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 number of micrographs collected for cryo-EM structure determination was determined by sample quality and microscope time availability, with the goal of obtaining the resolution structure possible. For the HEK293T radiotracer uptake experiments, sample sizes of at least n=3 (biologically independent replicates) were sufficient to ensure reproducibility. Predetermination of sample size was not needed for the studies reported here and thus no sample size calculation was performed.
Data exclusions	A couple datapoints were rejected (as indicated in Source Data file) based on technical issues. Junk particle removal via image classification is the underlying principle of cryo-EM data processing, therefore extensive image classification was used to to identify a subset of particle images that led to the highest resolution 3D-reconstruction.
Replication	At least triplicate (n ≥ 3) biological repeats were pursued for all functional experiments and were reproducible (sample size is noted in all cases in the figure legends).
Randomization	Randomization was not used for the functional and structural studies reported here because no grouping was needed.
Blinding	Blinding was not used for the functional and structural studies reported here since no grouping was performed.

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
☒	☐ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

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

Antibodies

Antibodies used	Monoclonal mouse anti-FLAG, clone M2 (Sigma-Aldrich, F3165) was used 1:1000 diluted. Polyclonal Donkey anti-mouse IgG (H+L) antibody conjugated to IRDye 800CW (LICOR, 926-32212) was used 1:5000 diluted.
Validation	From the manufacturer: Sensitivity Test Conforms: Detects 2 ng of FLAG-BAP fusion protein by dot blot using chemiluminescent detection. Specificity Confirmed: Detects a single band of protein on a western blot from an E. coli crude cell lysate.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Human embryonic kidney (HEK) 293S GnTI- cells and HEK293T cells are purchased from ATCC
Authentication	Cell lines were not authenticated.
Mycoplasma contamination	HEK293S GnTI- and HEK293T/17 cells were tested negative for mycoplasma contamination by Duke University Cell Culture Facility.

Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell line was used in this study.