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 <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

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| For | all st | atistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section. |
|-------------|-------------|---|
| n/a | Cor | nfirmed |
| | \boxtimes | The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement |
| X | | A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly |
| \boxtimes | | 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. |
| X | | A description of all covariates tested |
| X | | A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons |
| | \boxtimes | 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) |
| \boxtimes | | 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> |
| \boxtimes | | For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings |
| \boxtimes | | For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes |
| \boxtimes | | Estimates of effect sizes (e.g. Cohen's d, Pearson's r), indicating how they were calculated |
| | ' | Our web collection on <u>statistics for biologists</u> contains articles on many of the points above. |
| | | |

Software and code

Policy information about availability of computer code

Data collection

No software was used for data collection

Data analysis

For all structural biology data analysed in the context of this work, the following software were used: XDS v10.2022, STARANISO v3.350, PHASER v3.55.2, COOT v0.9.6, BUSTER v1.1.4, AlphaFold2 v2.3, Cryosparc v.4.2, Phenix v1.19-4092, APBS v1.4, CONSURF v3, DALI v5, UCSF Chimera X v1.0. All these tools are described in the literature and publicly available.

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 PabRpa1 AROD-OB-1 domains, PabRPA and PabRPA Tri-C/d20T crystal structures were deposited in the Protein Data Bank under accession codes, PDBid: 8AA9 (https://doi.org/10.2210/pdb8AA9/pdb), PDBid: 8AAJ (https://doi.org/10.2210/pdb8AAJ/pdb)

| AROD-OB-1 domains are doi.org/10.2210/pdb8C5 odb8OEL/pdb); EMDB-1 | doi.org/10.2210/pdb8AAS/pdb) respectively. The all-atom cryo-EM structure of PabRPA tetramer, as well as the 3D-class showing the deposited under accession code (PDBid: 8C5Y (https://doi.org/10.2210/pdb8C5Y/pdb); EMDB-16444) and (PDBid: 8C5Z (https://pdb); EMDB-16445) respectively. The cryo-EM maps of the condensed PabRPA/ssDNA complex (PDBid: 8OEL (https://doi.org/10.2210/827), relaxed PabRPA/ssDNA complex (PDBid: 8OEJ (https://doi.org/10.2210/pdb8OEJ/pdb); EMDB-16826). The individual PabRPA on EMDataResource under accession codes EMDB-16448 respectively. | | |
|---|--|--|--|
| Human resear | h participants | | |
| | t studies involving human research participants and Sex and Gender in Research. | | |
| Poporting on say an | gender N/A | | |
| Reporting on sex and | | | |
| Population characte | | | |
| Recruitment | N/A | | |
| Ethics oversight | N/A | | |
| Life sciences | elow that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection. Behavioural & social sciences Ecological, evolutionary & environmental sciences cument with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf | | |
| | es study design e on these points even when the disclosure is negative. | | |
| Sample size Bi | ding experiments that were performed in this work usually involve 4000 points. To determine the binding constants, binding experiments e performed at 7 different concentrations, and replicated as detailed in the figure legends (>=2). | | |
| Data exclusions No | data were excluded. | | |
| | ittempts at replication were succesfull. For example, for protein-protein interaction and protein-nucleic acids interaction measurements g biolayer interferometry, all experiments were succesfully reproduced at three times. | | |
| | binding studies, randomization of the data is not easily applicable. For an optimal binding experiments, the concentrations of the ligands at be selected in a concentration range that covers the dissociation constant. | | |
| ар | Blinding, which refers to the concealment of group allocation from one or more individuals involved in a clinical research study is not easily applicable to our biophysical experiments. Indeed, identifying the target protein and the control is important for the technical set-up of these experiments. For example, this enables us to perform the binding of the target protein at multiple concentrations simultaneously. | | |
| Reporting | or specific materials, systems and methods | | |
| · | om authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each materi 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 & exper | mental systems Methods | | |
| /a Involved in the s | n/a Involved in the study | | |

| Materials & experimental systems | | Methods | | |
|----------------------------------|-------------------------------|-------------|------------------------|--|
| n/a | Involved in the study | n/a | Involved in the study | |
| \boxtimes | Antibodies | \boxtimes | ChIP-seq | |
| \boxtimes | Eukaryotic cell lines | \boxtimes | Flow cytometry | |
| \boxtimes | Palaeontology and archaeology | \boxtimes | MRI-based neuroimaging | |
| \boxtimes | Animals and other organisms | | | |
| \boxtimes | Clinical data | | | |
| \boxtimes | Dual use research of concern | | | |
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