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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 <u>Authors & Referees</u> and the <u>Editorial Policy Checklist</u>.

Statistics

| 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 | 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) | | |
| x | | 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 Give <i>P</i> 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 a | bout <u>availability of computer code</u> |
|----------------------|--|
| Data collection | MS data collection: flexControl 3.4 (Bruker) |
| | Cryo-EM data collection: SerialEM, Focus |
| Data analysis | MS data analysis: flexAnalysis 3.3 (Bruker), SeeMS 3.0 |
| Data analysis | Cryo-EM data analysis and model building: RELION 3.0, CTFFind4, Chimera, PHENIX, COOT, Avogadro 1.2.0, PyMol 2.3 |

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. 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

Coordinates and charge density map were deposited in the RCSB Protein Data Bank with accession code 6U48 for the E. coli 50S ribosomal subunit in complex with PHZ. The source data (mass-spectra) underlying Supplementary Figs 1c, 2 and 3a,b are provided as Supplementary Data Files. Additional data are available from the corresponding author upon request.

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 <u>nature.com/documents/nr-reporting-summary-flat.pdf</u>

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

| Sample size | N/A |
|-----------------|---|
| Data exclusions | No data was excluded |
| Replication | MS measurements were performed at least in triplicate (both MALDI and ESI experiments). |
| Randomization | N/A |
| Blinding | N/A |

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 |
| × | Animals and other organisms |
| × | Human research participants |
| X | Clinical data |

Methods

| n/a | Involved in the study |
|-----|------------------------|
| × | ChIP-seq |
| × | Flow cytometry |
| × | MRI-based neuroimaging |