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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>.

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For	ali 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
	\boxtimes	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.
\boxtimes		A description of all covariates tested
\boxtimes		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 statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection

SerialEM (vs. 3.6)

Data analysis

IMOD (vs. 4.9.0), cisTEM (version 1.0-beta and pre-release), Frealign v. 9.11 (Aug 2017 release), EMAN(vs.2.07), RSRef(2000), Phenix (1.16-3549), Chimera (vs. 1.7), MacPyMOL: PyMOL v1.7.0.5 enhanced for MacOsX,

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 <u>data availability statement</u>. 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 models generated and analyzed during the current study will be available from the RCSB Protein Data Bank: 6WNW (unperturbed 70S ribosome), 6WNT (perturbed 50S subunit),6WNV (70S ribosome with the perturbed 50S subunit).

The cryo-EM maps used to generate models will be available from the Electron Microscopy Database: EMD-21858 (unperturbed 70S ribosome), EMD-21856 (perturbed 50S subunit), EMD-21857 (70S ribosome with the perturbed 50S subunit). The proteomic data are available in the PRIDE database under accession number PXD019058. The uncropped gels (Figs. 2f, 3a-c, Supplementary Figs 4c-d, and bar graph numeric data (Fig. 5c and Supplementary Fig. 2) are provided in the Source Data file.

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 & socia	al 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 dis	sclose on these points even when	the disclosure is negative.				
Sample size	All the proteomic and biochemical experiments were performed at least twice.					
Data exclusions	No data were excluded					
Replication	All the proteomic and biochemical experiments were performed at least twice. The rpesentative gels are shown in the relevant figures. Full uncropped gels are shown in the Source data file.					
Randomization	N/A					
Blinding	Blinding was impossible and irrelevant to the study due to the nature of biochemical experiments where the experimenter loads the gels/plates her/himself and then collects the data. However, the numeric nature of the data makes blinding irrelevant.					
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 Methods						
n/a Involved in th	,	n/a Involved in the study				
Antibodies		ChIP-seq				
Eukaryotic cell lines		Flow cytometry				
Palaeontology		MRI-based neuroimaging				
Animals an	Animals and other organisms					

Human research participants

Clinical data