

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 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 SerialEM-3.6.11

Data analysis MotionCor2-1.1.0, GCTF-1.0.6, Gautomatch-0.53, RELION 3.0, cryoSPARC-2.0, PHENIX-1.18rc1-3777, Coot-0.8.6, UCSF Chimera-1.10, Pymol-1.7.0.5, Chimera X- 1.2.5

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 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

The cryo-EM map of sGC in complex with YC1, riociguat and cinaciguat are accessible through the EMDB codes EMD-30619 [<https://www.ebi.ac.uk/pdbe/entry/emdb/EMD-30619>], EMD-30618 [<https://www.ebi.ac.uk/pdbe/entry/emdb/EMD-30618>], EMD-30620 [<https://www.ebi.ac.uk/pdbe/entry/emdb/EMD-30620>] and EMD-30621 [<https://www.ebi.ac.uk/pdbe/entry/emdb/EMD-30621>]. The atomic coordinates have been deposited in the Protein Data Bank (PDB) with accession codes 7D9S [<https://doi.org/10.2210/pdb7D9S/pdb>], 7D9R [<https://doi.org/10.2210/pdb7D9R/pdb>], 7D9T [<https://doi.org/10.2210/pdb7D9T/pdb>] and 7D9U [<https://doi.org/10.2210/pdb7D9U/pdb>]. PDB entries used in this study are available in the PDB database under accession codes 6JT1 [<https://doi.org/10.2210/pdb6JT1/pdb>], 6JT2 [<https://doi.org/10.2210/pdb6JT2/pdb>], 3L6J [<https://doi.org/10.2210/pdb3L6J/pdb>] and 5MNW [<https://doi.org/10.2210/pdb5MNW/pdb>].

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](https://www.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	All of functional experiments were repeated at least 3 times and the sample size is determined based on the reproducibility of the experiments.
Data exclusions	Cryo-EM micrographs with ice or ethane contamination, empty carbon, and poor CTF fit ($> 5 \text{ \AA}$) were excluded manually. Particles belonging to bad classes were discarded and the data processing flowchart were summarized in Supplementary Figures. These criteria were pre-established and the procedure is a common practise in cryo-EM image analysis.
Replication	All attempts at replication were successful according to the detailed protocol described in the methods section. The numbers of replication were described in figure legends.
Randomization	For cryo-EM 3D refinement, all particles were randomly split into two groups.
Blinding	The investigators were blinded to group allocation during cryo-EM data collection and analysis.

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 the study	n/a	Involved in the study
☒	☐ Antibodies	☒	☐ ChIP-seq
☐	☒ Eukaryotic cell lines	☒	☐ Flow cytometry
☒	☐ Palaeontology and archaeology	☒	☐ MRI-based neuroimaging
☒	☐ Animals and other organisms		
☒	☐ Human research participants		
☒	☐ Clinical data		
☒	☐ Dual use research of concern		

Eukaryotic cell lines

Policy information about [cell lines](#)

Cell line source(s)	Sf9 and sf21 were from Thermo Fisher Scientific.
Authentication	None of the cell line used was authenticated.
Mycoplasma contamination	All cell lines were tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.