

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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☒ | ☐ | 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
<i>Give P values as exact values whenever suitable.</i> |
| ☒ | ☐ | 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 EPU software V2.12 was used for data collection with Titan Krios G3i (Thermo Fisher).

Data analysis CryoSPARC v4.1.2, UCSF ChimeraX-1.9, Pymol v1.8.X, Coot 0.9.8.96, Phenix 1.21.2-5419, RELION 3.0, Topaz

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 model and maps of human MCM SH are deposited to PDB and EMDB with codes 9UQ0 and EMD-64408, respectively. The maps of human MCM SH States 2 and 3 are deposited to EMDB with codes EMD-68350 and EMD-68363, respectively. The model and maps of human MCM2-7Δ3CTE SH (State 1) are deposited to PDB and EMDB with codes 9VLN and EMD-65161, respectively. The model and maps of human MCM2-7Δ3CTE SH (State 2) are deposited to PDB and EMDB with codes 9VLW and EMD-65170, respectively. The model and map of human MCM2-7 (MCM3-Q761L) are deposited to PDB and EMDB with codes 22VT and EMD-68728,

respectively. Source data are provided with this paper.

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender N/A

Reporting on race, ethnicity, or other socially relevant groupings N/A

Population characteristics N/A

Recruitment N/A

Ethics oversight N/A

Note that full information on the approval of the study protocol must also be provided in the manuscript.

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 No statistical methods were used to predetermine sample size. All biochemical experiments were replicated two or three times. For structural data, the number of micrographs collected was determined based on the target resolution of the relevant electron microscopy 3D structures.

Data exclusions Regarding the cryo-EM raw micrograph screening, exclusion was done based on the quality of the images. Regarding the particle selection, 2D and 3D classification were used and criterion is based on the quality of resulting 2D class average and 3D maps.

Replication All attempts of replication were successful. Cryo-EM single particle analysis relies on averaging over a large number of independent observations.

Randomization Samples were not allocated to groups.

Blinding Investigators were not blinded during data acquisition and analysis because because visual inspection is necessary for the methods employed.

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

☒ ☐ Clinical data

☒ ☐ Dual use research of concern

☒ ☐ Plants

Methods

n/a Involved in the study

☒ ☐ ChIP-seq

☐ ☒ Flow cytometry

☒ ☐ MRI-based neuroimaging

Antibodies

Antibodies used	anti-FLAG (1:2000, Sigma, F1804), anti-MCM2 (1:2000, Bethyl, A300-122A), anti-MCM3 (1:1000, Santa Cruz, sc-390480), anti-MCM4 (1:2000, Bethyl, A300-193A), anti-MCM5 (1:2000, Bethyl, A300-195A), anti-MCM6 (1:2000, Bethyl, A300-194A), anti-MCM7 (1:500, Santa Cruz, sc-9966), anti-Vinculin (1:1000, Santa Cruz, sc-73614), anti- β -Actin (1:5000, Cell Signaling Technology, 4970), anti-H2B (1:8000, Abcam, ab1790), anti- γ H2A.X phospho S139 (1:2000, Abcam, ab2893), anti-strep (1:8000, Abcam, ab307676)
Validation	All primary antibodies are commercially available and validated by the manufacturers.

Eukaryotic cell lines

Policy information about [cell lines and Sex and Gender in Research](#)

Cell line source(s)	HeLa S3, HCT116, and HEK 293T were obtained from ATCC. HCT116 mAID-MCM3 was a gift from Masato Kanemaki.
Authentication	The cells were used only for protein purification and functional assays, and not further authenticated.
Mycoplasma contamination	Cell lines were not tested for mycoplasma contamination
Commonly misidentified lines (See ICLAC register)	N/A

Plants

Seed stocks	N/A
Novel plant genotypes	N/A
Authentication	N/A

Flow Cytometry

Plots

Confirm that:

- ☐ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).
- ☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).
- ☐ All plots are contour plots with outliers or pseudocolor plots.
- ☐ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	Cells were fixed in 75% ice-cold ethanol for ≥ 30 min. After washing twice with PBS, cells were resuspended in FACS buffer (50 μ g/mL propidium iodide, 100 μ g/mL RNase A, 0.1% Triton X-100 in PBS) and incubated in the dark for 30 min.
Instrument	BD FACSAria™ III Cell Sorter
Software	FlowJo™ v10.4
Cell population abundance	N/A
Gating strategy	Gating Strategy is provided in Supplementary Figure 10

- ☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.