nature portfolio

Corresponding author(s): Weixing Zhao

Last updated by author(s): Jul 17, 2024

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 *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	Not applicable
Data analysis	AlphaFold2 for predicting potential conformations of the BRCA2-DBD-DSS1 complex; FlowJo software for Flow cytometry analsysis; Prism 10-GraphPad Software for all the statistic data analysis.

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

Provide your data availability statement here.

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	Not applicable
Reporting on race, ethnicity, or other socially relevant groupings	Not applicable
Population characteristics	Not applicable
Recruitment	Not applicable
Ethics oversight	Not applicable

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

Life sciences study design

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

Sample size	No sample size calculation was performed. We have used the data from at least three independent experiments. Three repeats is usually a good starting place for evaluating the spread of the data. Importantly, the P values results from statistical analyses suggested that the samples sizes in our study are sufficient.
Data exclusions	Not applicable
Replication	The experimental findings were reliably reproduced and the data are analyzed from at least three independent experiments.
Randomization	Not Applicable
Blinding	Blinding was not part of the study design. The research involved in molecular analysis of specific proteins and the researchers needed to produce and characterize them and their truncations or domains.

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	For Immunoblot analysis: DSS1(SC28848, Santa Cruz; 1:500), BRCA2 (EMD Millipore, OP95-100UG; 1:1000), RAD51 (Novus Biologicals, NB100-148; 1:4000), Tubulin (2128S, Cell Signaling; 1:2000), GAPDH (2118S, Cell Signaling; 1:15000), Lamin B1

(SC374015, Santa Cruz, 1:500), RPA70 (Abcam ab79398; 1:3000), p-RPA32(S4/S8, A300-245A, Bethyl Laboratories; 1:2000), Phospho-Histone gH2AX (9718S, Cell Signaling; 1:2000), Flag M2-HRP (Sigma, A8592; 1:2000), GST-HRP (Invitrogen, MA4-004-HRP; 1:4000), His-HRP (Sigma, A7058, 1:3000), RPA32 (Abcam, ab2175; 1:2000), RPA70 (2267S, Cell Signaling; 1:2000) and Actin (Abcam, ab3280; 1:3000), HRP-conjugated secondary antibodies (Pierce 31450 for rabbit anti-mouse IgG-HRP; Sigma A6154 for goat anti-rabbit IgG-HRP; Santa Cruz Biotech SC2032 for goat anti-rat IgG-HRP). For Immunofluorescence staining : RAD51(8875S, Cell Signaling; 1:500), gH2AX (05636, Millipore; 1:500), RPA (MABE285, Millipore, 1:500) and BRCA2 (Homemade from Xia Bing lab).

Validation

The primary antibodies were validated for use based on the position the antigen in the SDS-PAGE gels and the disappearance upon siRNA knock down in this study.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	HEK293T (ATCC), HeLa (ATCC) and U2OS clones (DR-GFP; a gift from Jeremy Stark)
Authentication	The cell lines used have not been authenticated by us.
Mycoplasma contamination	All the cell lines were tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

Plants

Seed stocks	Not Applicable
Novel plant genotypes	Not Applicable
Authentication	Not Applicable

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	U2OS Cells were trypsinized and suspended in DPBS buffer.
Instrument	BD FACSCalibur S
Software	We run our experiments with CellQuest Pro and analyze the data with Flowjo V10
Cell population abundance	All single live cells were used for analysis
Gating strategy	Untreated cells(GFP-negative) were used for gating and GFP-positive single cells were counted in the single live cell population.

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