nature portfolio

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Last updated by author(s): Oct 15, 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	LiCor Image Acquisition Software 1.1, SoftMax Pro 7.1, Leica Application Suite 3.5.5.19976
Data analysis	LiCor ImageStudioLite 5.0, CellProfiler 4.2.5, GraphPad Prism 10.3

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 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

Source data are provided with this paper as a Source data file.

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	Our experiments are performed on common, asexually reproducing cell cultures, and therefore sex and gender are not relevant to our study.
Reporting on race, ethnicity, or other socially relevant groupings	Our experiments are performed on cell cultures, and therefore social factors do not apply.
Population characteristics	No human participants were employed in this study.
Recruitment	No human participants were employed in this study.
Ethics oversight	No human participants were employed in this study.

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 statistical method was used to predetermine sample size. Required sample sizes were estimated on standard practices in the lab for each given method, and increased or decreased based on variance in preliminary experiments to distribute resources most efficiently.
Data exclusions	No data were excluded.
Replication	Experiments were performed by multiple independent operators on different days in different locations. Due to the natural growth of cell lines, experiments were replicated in cells with different passage numbers. Experiments were also replicated in different cell lines, as reported in the manuscript.
Randomization	All experiments are internally controlled. Each experiment is carried out on multiple wells derived two days prior from the same stock of cells. Therefore, randomization is naturally achieved by passaging the cells. It is not possible to randomize the conditions across plates because what differs between wells is the temporal patterning of treatments, and those must be precisely allocated to specific wells to make it possible to perform the protocol in real time.
Blinding	Due to reasons explained above, it is not possible to blind experimenters to the conditions of the experiment — all the temporal patterning is done manually. The experimenters were blind to the conditions of the experiment when analyzing Western blots and immunofluorescence images.

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-P-ERK (CST #9101, polyclonal, 1:1000, lot 32), anti-ERK (CST #4696, L34F12, 1:1000, lot 33), anti-P-CREB (CST #9198, 87G3, 1:1000, lot 19), anti-CREB (CST #9104, 86B10, 1:1000, lot 8), anti-firefly luciferase (Invitrogen PA5-32209, polyclonal, 1:1000, lot ZF4348385), anti-β-actin (CST #3700, 8H10D10, 1:5000, lot 21), anti-H3 histone (CST #4499, D1H2, 1:10,000, lot 9)
Validation	P-ERK (CST #9101: https://www.cellsignal.com/products/primary-antibodies/phospho-p44-42-mapk-erk1-2-thr202-tyr204-antibody/9101), ERK (CST #4696: https://www.cellsignal.com/products/primary-antibodies/p44-42-mapk-erk1-2-l34f12-mouse-mab/4696), P-CREB (CST #9198: https://www.cellsignal.com/products/primary-antibodies/phospho-creb-ser133-87g3-rabbit-mab/9198), CREB (CST #9104: https://www.cellsignal.com/products/primary-antibodies/creb-86b10-mouse-mab/9104), firefly luciferase (Invitrogen PA5-32209: https://www.thermofisher.com/antibody/product/Firefly-luciferase-Antibody-Polyclonal/PA5-32209), β-actin (CST #3700: https://www.cellsignal.com/products/primary-antibodies/b-actin-8h10d10-mouse-mab/3700), H3 histone (CST #4499: https://www.cellsignal.com/products/primary-antibodies/histone-h3-d1h2-xp-rabbit-mab/4499)

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	SH-SY5Y: ATCC, cat no. CRL-2266; HEK-293: ATCC, cat no. CRL-1573
Authentication	Cell lines were obtained directly from ATCC and not further authenticated.
Mycoplasma contamination	Cell lines were not tested for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	HEK-293 is occasionally misidentified with HeLa. Our reviwer requested validation of our data in one of these two cell lines.

Plants

Seed stocks	No plants were used in this study.
Novel plant genotypes	No plants were used in this study.
Authentication	No plants were used in this study.