# nature portfolio

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# 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 | Python code for automated image acquisition. |
| Data analysis | FiJi ImageJ (ver. 1.53t), ImageJ plugin RankFilters (https://imagej.nih.gov/ij/source/ij/plugin/filter/RankFilters.java , ver. 2020-07-17), Living Image (ver. 4.5.5), Tecan SPARKCONTROL Dashboard (ver. 3.1 SP1), FusionCapt Advance Pulse 7 (ver. 17.03), LibRaw (ver. 0.19.2), custom Python code for image and data processing (Python ver. 3.10.12) (pandas (https://pandas.pydata.org/ , ver. 1.5.1) package, Seaborn (https://seaborn.pydata.org/ , ver. 0.12.2) package, matplotlib (https://matplotlib.org/ , ver. 3.7.1) package, numpy (https://numpy.org/ , ver. 1.23.5) package, scipy package (https://www.scipy.org/ , ver. 1.11.3). Scikit-posthocs Python package (https://pypi.org/project/scikit-posthocs/, ver. 0.8.0) for statistical 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](#)

Data is available at https://doi.org/10.6084/m9.figshare.24623817. Python code for processing and plotting data is available at both https://github.com/Perfus/BL2.0 and https://doi.org/10.6084/m9.figshare.24623976.

## Human research participants

Policy information about [studies involving human research participants and Sex and Gender in Research.](#)

| Reporting on sex and gender | 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](#)

# Life sciences study design

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

| Sample size | The experiments described in this study were done for the first time. Due to exploratory nature of our study we refrained from unnecessary generalizations. No pre-specified effect size could be determined a priori. |
| Data exclusions | No data were excluded from the study. |
| Replication | Number of replicates are explicitly stated in the figure legends. Where applicable, reported results were consistently replicated across multiple experiments with all replicates generating similar results. |
| Randomization | No randomisation was performed. Our experiments had measurable and observable outcomes (e.g., enzyme activity, light emission etc). These outcomes are not influenced by subjective bias, making randomisation less relevant. |
| Blinding | No blinding was performed. Our experiments had measurable and observable outcomes (e.g., enzyme activity, light emission etc). These outcomes are not influenced by subjective bias, making blinding less relevant. |

# 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 |

## Methods

| n/a | Involved in the study |
|---|---|
| ☒ | ☐ ChIP-seq |
| ☒ | ☐ Flow cytometry |
| ☒ | ☐ MRI-based neuroimaging |

## Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

| | |
|---|---|
| Cell line source(s) | HEK293 (ATCC), HEK293NT (unknown origin) |
| Authentication | Cell line was authenticated morphologically. |
| Mycoplasma contamination | Cell lines were frequently tested for mycoplasma contamination. Cell line used in this study was verified to be mycoplasma negative before undertaking experiments with it. |
| Commonly misidentified lines (See ICLAC register) | No commonly misidentified cells were used. All cells displayed homogeneous characteristic morphology. |