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Life Sciences Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form is intended for publication with all accepted life science papers and provides structure for consistency and transparency in reporting. Every life science submission will use this form; some list items might not apply to an individual manuscript, but all fields must be completed for clarity.

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Please do not complete any field with "not applicable" or n/a. Refer to the help text for what text to use if an item is not relevant to your study.
For final submission: please carefully check your responses for accuracy; you will not be able to make changes later.

Experimental design

1. Sample size

   Describe how sample size was determined. — All imaging has been done on single cells.

2. Data exclusions

   Describe any data exclusions. — Cells have been selected visually according to staining quality.

3. Replication

   Describe the measures taken to verify the reproducibility of the experimental findings. — We repeated several single cell imaging (>10 typically) for ensuring the reproducibility of the experimental findings.

4. Randomization

   Describe how samples/organisms/participants were allocated into experimental groups. — This is not applicable for single cell imaging.

5. Blinding

   Describe whether the investigators were blinded to group allocation during data collection and/or analysis. — This is not applicable for single cell imaging.

Note: all in vivo studies must report how sample size was determined and whether blinding and randomization were used.

6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

n/a	Confirmed
☐	☒ The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)
☐	☒ A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☒	☐ A statement indicating how many times each experiment was replicated
☒	☐ 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 any assumptions or corrections, such as an adjustment for multiple comparisons
☒	☐ Test values indicating whether an effect is present *Provide confidence intervals or give results of significance tests (e.g. P values) as exact values whenever appropriate and with effect sizes noted.*
☐	☒ A clear description of statistics including central tendency (e.g. median, mean) and variation (e.g. standard deviation, interquartile range)
☒	☐ Clearly defined error bars in all relevant figure captions (with explicit mention of central tendency and variation)

See the web collection on statistics for biologists for further resources and guidance.

Software

Policy information about availability of computer code

7. Software

Describe the software used to analyze the data in this study.	We used Image J 1.51k, Matlab 2015a, 2017b (Mathworks) and Imaris (Bitplane). Custom algorithms for quantitative phase reconstruction and for 3D bSOFI analysis were implemented in Matlab (see Supplementary Material for description).

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). *Nature Methods* guidance for providing algorithms and software for publication provides further information on this topic.

Materials and reagents

Policy information about availability of materials

8. Materials availability

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a third party.	All unique materials are readily available upon request from the authors or from commercial source.

9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).	anti alpha-tubulin DM1a mouse monoclonal, (ref ab7291, Abcam, validated by Abcam), anti-pS129-α-syn rabbit (ref ab168381, clone MJFR-13, Abcam) anti-α-syn mouse (ref 610787, clone SYN-1, BD) anti-MAP2 chicken (ref ab92434, Abcam) anti-donkey anti-mouse IgG (H+L) highly cross-adsorbed secondary antibody, Alexa 647 Fluor (A-31571, Invitrogen, validated by Invitrogen). donkey anti-rabbit Alexa647 IgG (H+L) highly cross-adsorbed secondary antibody(A-31573, Invitrogen, validated by Invitrogen) donkey anti-mouse Alexa568 IgG (H+L) highly cross-adsorbed secondary antibody(A10037, Invitrogen, validated by Invitrogen) Alexa Fluor® 488 AffiniPure Donkey Anti-Chicken IgY (IgG) (H+L) (Jackson ImmunoResearch)

10. Eukaryotic cell lines

a. State the source of each eukaryotic cell line used.	RAW 264.7 cells were a gift from the lab of B. Deplancke (LSBG-EPFL), fibroblast cells were a gift from M. Ricchetti from Institute Pasteur, HeLa cells were obtained from ATCC, and HEK293T cells were a gift from the lab of A. Radenovic (LBEN-EPFL).
b. Describe the method of cell line authentication used.	We did not perform authentication of the cell lines.
c. Report whether the cell lines were tested for mycoplasma contamination.	Cell lines were not tested for mycoplasma contamination.
d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by ICLAC, provide a scientific rationale for their use.	No commonly misidentified cell lines were used.

Animals and human research participants

Policy information about studies involving animals; when reporting animal research, follow the ARRIVE guidelines

11. Description of research animals

Provide all relevant details on animals and/or animal-derived materials used in the study.	Pregnant female C57BL/6J Rcc Hsd were obtained from Harlan Laboratories (France) and were housed according to the Swiss legislation and the European Community Council directive (86/609/EEC). Primary hippocampal cultures were prepared from mice brains from P0 pups as described previously (see Methods section)

Policy information about studies involving human research participants

12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.	The study did not involve human participants.