

Corresponding author(s): NCOMMS-18-32535B

Reporting Summary

Nature Research 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 Research policies, see <u>Authors & Referees</u> and the <u>Editorial Policy Checklist</u>.

When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. figure legend, table legend, main

Statistical parameters

text	, or N	Methods section).	
n/a	Confirmed		
	\boxtimes	The $\underline{\text{exact sample size}}(n)$ for each experimental group/condition, given as a discrete number and unit of measurement	
	\boxtimes	An indication of 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.	
X		A description of all covariates tested	
	\boxtimes	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons	
	\boxtimes	A full description of the statistics including <u>central tendency</u> (e.g. means) or other basic estimates (e.g. regression coefficient) AND <u>variation</u> (e.g. standard deviation) or associated <u>estimates of uncertainty</u> (e.g. confidence intervals)	
\boxtimes		For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>	
\times		For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings	
X		For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes	
\boxtimes		Estimates of effect sizes (e.g. Cohen's d, Pearson's r), indicating how they were calculated	
	\boxtimes	Clearly defined error bars State explicitly what error bars represent (e.a. SD. SF. Cl.)	

Our web collection on <u>statistics for biologists</u> may be useful.

Software and code

Policy information about availability of computer code

Data collection

All the images with microscopy were collected with Zen software (Carl Zeiss)

Western blot images were collected with LAS-1000 (FujiFilm)

Data analysis

Colocalization between injected liposomes and endogenous organelles were analyzed with a Matlab algorithm provided by Prof. Silvio Rizzoli. Fluorescence intensity and perinuclear index was analyzed with Image J software.

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/reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a <u>data availability statement</u>. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data

- A description o	f any restrictions on data availability				
The plasmids and experimentally data that support the findings of this study are available from the corresponding authors upon reasonable request.					
Field-spe	cific reporting				
Please select the be	est fit for your research. If you are not sure, read the appropriate sections before making your selection.				
\times Life sciences	Behavioural & social sciences Ecological, evolutionary & environmental sciences				
For a reference copy of t	he document with all sections, see <u>nature.com/authors/policies/ReportingSummary-flat.pdf</u>				
Life scier	nces study design				
All studies must dis	close on these points even when the disclosure is negative.				
Sample size	The number of independent experiments used for the calculation of mean values and S.E.M. values is given in the legend to each panel. No statistical methods were applied to predetermine sample size.				
Data exclusions	No data were excluded.				
Replication	Experiments were repeated at least three independent experiments. If there were outliers, experiments were performed more to reduce S.E.M				
Randomization	no applicable				
Blinding	Not applied in this study				
Reportin	g for specific materials, systems and methods				
reportin	8 101 3 peem 6 11 atemais, 3 y sterms and 11 et 10 as				
Materials & experimental systems Methods					
n/a Involved in the study n/a Involved in the study					
	logical materials ChIP-seq				
Antibodies					
Eukaryotic cell lines MRI-based neuroimaging Palaeontology					
Animals and other organisms					
Human res	earch participants				

Antibodies

Antibodies used

Primary Antibodies

Anti-APPL1 (Rabbit monoclonal, #3858, Cell Signaling Technology) anti LC3B (Rabbit monoclonal, #4599, Cell Signaling Technology) anti-Rab7 (Rabbit monoclonal, #9367, Cell Signaling Technology)

anti-EEA1 (Mouse monoclonal, clone 14, #612006, Becton, Dickinson)

anti-GM130 (Mouse monoclonal, clone 35, #560257, Becton, Dickinson)

anti-M6PR (Mouse monoclonal, clone 2G11, ab2733, Abcam)

anti-LAMP1 (Rabbit polyclonal, ab24170, Abcam)

anti-mitofilin (Mouse monoclonal, clone 2E4AD5, ab110329, Abcam)

anti-mitofusion2 (Rabbit polyclonal, ab101055, Abcam)

anti-cathepsin D (Rabbit polyclonal, ab6313, Abcam)

anti-transferrin receptor (Mouse monoclonal, clone H68.4, sc-65882, Santa Cruz Biotechnology)

```
anti-LBPA (Mouse monoclonal, clone 6C4, 7-PLBPA, Echelon)
anti-Golgin97 (Mouse monoclonal, clone CDF4, A-21270, Thermo Fisher Scientific)
anti-RFP (Rabbit polyclonal, R10367, Thermo Fisher Scientific)
anti-Vps52 (Rabbit polyclonal, PA5-24408, Thermo Fisher Scientific)
anti-Rab11 (Rabbit polyclonal, 71-5300, Thermo Fisher Scientific)
anti-PDI (Mouse monoclonal, clone 7H25L2, 700782, ABfinity)
anti-Vps51 (Rabbit polyclonal, HPA061447, Atlas antibodies)
anti-Vps13B (Rabbit polyclonal, HPA043865, Atlas antibodies)
anti-CD63 (Mouse monoclonal, clone H5C6, Developmental Studies Hybridoma Bank)
anti-Rab6 (Rabbit polyclonal, 10187-2-P, Proteintech)
anti-Rab35 (Rabbit polyclonal,11329-2-AP ,Proteintech)
anti-Rab14 (Rabbit polyclonal, R0656, Sigma-Aldrich)
anti-GFP (Rabbit polyclonal, 132002, Synaptic system)
anti-α-tubulin (Mouse Monoclonal, clone 3A2, 302211, Synaptic system)
anti-beta-actin (Rabbit polyclonal, 251003, Synaptic system)
anti-Rab5 (Mouse Monoclonal, clone 621.3, 108011, Synaptic system)
anti-syntaxin 6 (Rabbit polyclonal, 110062, Synaptic system)
anti-syntaxin 13 (Rabbit polyclonal, 110132, Synaptic system)
anti-syntaxin 4 (Rabbit polyclonal, 110042, Synaptic system)
anti-syntaxin 16 (Rabbit polyclonal, 110162, Synaptic system)
anti-VAMP4 (Rabbit polyclonal, 136002, Synaptic system)
Secondary antibodies
Alexa Fluor 488-conjugated goat anti-mouse (115-545-166, Jackson ImmunoResarch Laboratories)
Cy3-conjugated goat anti-mouse (115-165-146, Jackson ImmunoResarch Laboratories)
Cy5-conjugated goat anti-mouse (115-175-166, Jackson ImmunoResarch Laboratories)
HRP-conjugated goat anti-mouse (STAR117P, BioRad)
Alexa Fluor 488-conjugated goat anti-rabbit (111-545-144, Jackson ImmunoResarch Laboratories)
Cy3-conjugated goat anti-rabbit (111-165-144, Jackson ImmunoResarch Laboratories)
Cy5-conjugated goat anti-rabbit (111-175-144, Jackson ImmunoResarch Laboratories)
HRP-conjugated goat anti-rabbit (5196-2504, BioRad)
```

Validation

```
Anti-APPL1 (Rabbit polyclonal, #3858, Cell Signaling Technology, https://www.cstj.co.jp/search/index.php?
queryString=appl1&x=0&v=0)
anti LC3B (#4599, Cell Signaling Technology, https://www.cstj.co.jp/products/4599.html)
anti-Rab7 (#9367, Cell Signaling Technology, https://www.cstj.co.jp/products/9367.html)
anti-EEA1 (#612006, Becton, Dickinson, https://www5.bdj.co.jp/reagents/view/610456)
anti-GM130 (#560257, Becton, Dickinson, https://www5.bdj.co.jp/reagents/view/610822)
anti-M6PR (ab2733, Abcam, https://www.abcam.co.jp/m6pr-cation-independent-antibody-2g11-ab2733.html)
anti-LAMP1 (ab24170, Abcam, https://www.abcam.co.jp/lamp1-antibody-lysosome-marker-ab24170.html)
anti-mit of ilin\ (ab 110329, Abcam, \ https://www.abcam.co.jp/mit of ilin-antibody-2e4ad5-mit ochondrial-marker-ab 110329. html)
anti-mitofusion2 (ab101055, Abcam, https://www.abcam.co.jp/mitofusin-2-antibody-ab101055.html)
anti-cathepsin D (ab6313, Abcam, https://www.abcam.co.jp/cathepsin-d-antibody-ctd-19-ab6313.html)
anti-transferrin receptor ( sc-65882, Santa Cruz Biotechnology, https://www.scbt.com/scbt/ja/product/cd71-antibody-h68-4?
requestFrom=search)
anti-LBPA (Z-PLBPA, Echelon, http://www.echelon-inc.com/index.php?module=Products&func=detail&id=767)
anti-Golgin97 (A-21270, Thermo Fisher Scientific, https://www.thermofisher.com/antibody/product/Golgin-97-Antibody-clone-
CDF4-Monoclonal/A-21270)
anti-RFP (Rabbit polyclonal, R10367, Thermo Fisher Scientific, https://www.thermofisher.com/antibody/product/RFP-Tag-
Antibody-Polyclonal/R10367)
anti-Vps52 (PA5-24408, Thermo Fisher Scientific, https://www.thermofisher.com/antibody/product/VPS52-Antibody-Polyclonal/
PA5-24408)
anti-Rab11 (Rabbit polyclonal, 71-5300, Thermo Fisher Scientific, https://www.thermofisher.com/antibody/primary/target/
rab11)
anti-PDI (700782, ABfinity, https://www.thermofisher.com/antibody/product/PDIA2-Antibody-clone-7H25L2-
Monoclonal/700782)
anti-Vps51 (HPA061447, Atlas antibodies, https://atlasantibodies.com/products/VPS51-antibody-HPA061447)
anti-Vps13B (HPA043865, Atlas antibodies, https://atlasantibodies.com/products/VPS13B-antibody-HPA043865)
anti-CD63 (Mouse monoclonal, clone H5C6, Developmental Studies Hybridoma Bank, http://dshb.biology.uiowa.edu/H5C6)
anti-Rab6 (10187-2-P, Proteintech, https://www.ptglab.com/products/RAB6A-Antibody-10187-2-AP.htm)
anti-Rab35 (11329-2-AP ,Proteintech, https://www.ptglab.com/Products/RAB35-Antibody-11329-2-AP.htm)
anti-Rab14 (R0656, Sigma-Aldrich, https://www.sigmaaldrich.com/catalog/product/sigma/r0656?lang=de&region=DE)
anti-GFP (132002, Synaptic system, https://www.sysy.com/products/gfp/facts-132002.php)
anti-α-tubulin (clone 3A2, 302211, Synaptic system, https://www.sysy.com/products/tubulin/facts-302211.php)
anti-beta-actin (Rabbit polyclonal, 251003, Synaptic system, https://www.sysy.com/products/b-actin/facts-251003.php)
anti-Rab5 (108011, Synaptic system, https://www.sysy.com/products/rab5/facts-108011.php)
anti-syntaxin 6 (110062, Synaptic system, https://www.sysy.com/products/syntaxin6/facts-110062.php)
anti-syntaxin 13 (110132, Synaptic system, https://www.sysy.com/products/syntaxin13/facts-110132.php)
anti-syntaxin 4 (110042, Synaptic system, https://www.sysy.com/products/syntaxin4/facts-110042.php)
anti-syntaxin 16 (110162, Synaptic system, https://www.sysy.com/products/syntaxin16/facts-110162.php)
anti-VAMP4 (136002, Synaptic system, https://www.sysy.com/products/vamp4/facts-136002.php)
```

Eukaryotic cell lines

Policy information about <u>cell lines</u>	
Cell line source(s)	HeLa cell, Vero cell
Authentication	none
Authentication	none
Mycoplasma contamination	The cell lines were not tested for Mycoplasma contamination
Commonly misidentified lines	none
(See <u>ICLAC</u> register)	