

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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☒ | ☐ | 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
<i>Give P values as exact values whenever suitable.</i> |
| ☒ | ☐ | 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

We used the autoPROC software to process and scale the X-ray diffraction data. We used the UNICORN 6 to collect analytic gel filtration chromatography data. We used the ASTRA 6 to collect multi-angle light scattering data. We used the Agilent VnmrJ 3.2 to collect NMR data. We used the SoftMax Pro 7.0 to collect fluorescence polarization data. We used the automated ITC calorimeter (Malvern) to collect the ITC data.

Data analysis

We used the PHENIX 1.20, Coot 0.9.6, MolProbity, and Pymol 2.5.2 to determine and analyse the two crystal structures reported in our manuscript. We used Origin9 to analyse the analytic gel filtration chromatography data. We used GraphPad Prism 9 to analyse fluorescence polarization data. We used the NMRDraw 8.1, Sparky 3.115 and CCPN to analyse the NMR data. We used ImageJ2 2.14.0 and GraphPad Prism 9 to analyse the Western Blotting data. We used the Malvern MicroCal PEAQ-ITC Analysis Software 1.1.0.1262 to analyse the ITC data.

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](#)

The coordinates and structure factors of the FIP200 Claw/ATG16L1 FIR complex and the GABARAPL1/ATG16L1 FIR complex solved in this study have been deposited in the Protein Data Bank under the accession code 9J54 and 9JF2, respectively. Source data are provided as a Source Data file. All additional experimental data are available from the corresponding author on request.

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	N/A
Reporting on race, ethnicity, or other socially relevant groupings	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](https://www.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 determine the correct sample size. Instead, the sample size was chosen on the basis of previous experience with each specific assay. Most assays are highly reproducible when experiments are repeated on multiple occasions.
Data exclusions	No data was excluded from the study.
Replication	All biological experiments were carried out under clearly defined and standard conditions and were repeated at least twice whenever possible. All replication attempts were successful.
Randomization	The samples were randomly allocated.
Blinding	Investigators were not blinded during data collection and analysis as experimental setup and analyses were performed by the same individuals.

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-GFP (Proteintech, 50430-2-AP, 1:1000 dilution) anti-GFP (Proteintech, 66002-1-Ig, 1:2000 dilution) anti-GFP (Abmart, M20004M, 1:2000 dilution) anti-Flag (Proteintech, 20543-1-AP, 1:1000 dilution) anti-Flag (Proteintech, 66008-4-Ig, 1:2000 dilution) anti-ATG16L1 (Abcam, ab187671, 1:2000 dilution) anti-LC3B (Abcam, ab192890, 1:2000 dilution) anti- β -actin (Proteintech, 66009-1-Ig, 1:5000 dilution) anti-p62 antibody (Cell Signaling Technology, 39749S, 1:2000 dilution) anti-FIP200 (Proteintech, 17250-1-AP, 1:2000 dilution) anti-ATG5 (Proteintech, 81803-1-RR, 1:2000 dilution) anti-WIP1 (Proteintech, 28820-1-AP, 1:2000 dilution) anti-GABARAP family (Selleck, F1156, 1:2000 dilution)
Validation	All antibodies were purchased from commercial vendors who provide validation information on their websites.

Eukaryotic cell lines

Policy information about [cell lines and Sex and Gender in Research](#)

Cell line source(s)	HEK293T and HeLa cell lines were kindly provided by Prof. Junying Yuan from Interdisciplinary Research Center on Biology and Chemistry, CAS, Shanghai, China.
Authentication	Authentication was not performed as none of the cells used have been listed in the commonly misidentified lines.
Mycoplasma contamination	Cell lines were regularly tested for mycoplasma using the MycAway TM -Color One-Step Mycoplasma Detection Kit (YEASEN M17371).
Commonly misidentified lines (See ICLAC register)	No misidentified lines were used in this study.

Plants

Seed stocks	N/A
Novel plant genotypes	N/A
Authentication	N/A