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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, seeAuthors & Referees and theEditorial Policy Checklist.

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| For all s | statistical analys | ses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section. | | | | |
|-----------|--|--|--|--|--|--|
| n/a Co | onfirmed | | | | | |
| x | The exact san | nple size (n) for each experimental group/condition, given as a discrete number and unit of measurement | | | | |
| | A statement of | on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly | | | | |
| x | | stical test(s) used AND whether they are one- or two-sided mon tests should be described solely by name; describe more complex techniques in the Methods section. | | | | |
| x | A description | of all covariates tested | | | | |
| × | A description | of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons | | | | |
| x | A full descript AND variation | cion of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals) | | | | |
| × | | othesis testing, the test statistic (e.g. F , t , r) with confidence intervals, effect sizes, degrees of freedom and P value noted 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 <u>statistics for biologists</u> contains articles on many of the points above. | | | | |
| Softv | ware and o | code | | | | |
| Policy ir | nformation abo | ut <u>availability of computer code</u> | | | | |
| Data | collection | All data collection described in Methods. All immunofluorescence images and movies collected via NIS Elements AR 5.2 (Nikon). | | | | |

Data analysis

Microscopy images were processand analyzed using NIS-element software (Nikon Corp., Tokyo, Japan), ImageJ or Volocity software. For statistical analysis GraphPad Prism software was used. Software used for crystal structure determination and analysis are described in the

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. 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 of any restrictions on data availability

The atomic coordinates and structure factors of B2-HpComF have been deposited at the Brookhaven Protein Data Bank under the accession number 7P0H. When not present in the main text or the supplementary materials, data that support the findings are available from the correponding authors upon reasonable request.

| Field-spe | ecific reporti | ng | | | |
|--|------------------------------------|---|--|--|--|
| Please select the o | ne below that is the best f | it for your research. If you are not sure, read the appropriate sections before making your selection. | | | |
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| Life scier | nces study d | esign | | | |
| | | n when the disclosure is negative. | | | |
| Sample size | | le sizes were deStaeirsmisisnezddauccteoxrdteinigcito commonly accepted standards in the field. The number of independent iments and the number of cells scored (n) are indicated in the figures and figure legends. | | | |
| Data exclusions | n/a | | | | |
| Replication | As mentioned above experi | oned above experiments were repeated and were only discarded when the controls failed. | | | |
| Randomization | n/a | | | | |
| Blinding | n/a | | | | |
| | | | | | |
| Reportin | g for specifi | c materials, systems and methods | | | |
| We require informati | ion from authors about some | types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, If you are not sure if a list item applies to your research, read the appropriate section before selecting a response. | | | |
| Materials & experimental systems | | Methods | | | |
| n/a Involved in the study | | n/a Involved in the study | | | |
| Antibodies | | X ChIP-seq | | | |
| Eukaryotic cell lines | | Flow cytometry | | | |
| Palaeontology MRI-based neuroimaging Animals and other organisms | | | | | |
| | search participants | | | | |
| Clinical da | ta | | | | |
| A ±1111 | | | | | |
| <u>Antibodies</u> | | | | | |
| Antibodies used | | AG Monoclonal Sigma F1804-1MG 088K6018; Rabbit anti HpComF produced in our laboratory; Rabbit anti-MotB Boneca's laboratory (Institut Pasteur) | | | |

The anti-ComF antibody was validated using extracts from bacterial strains mutated in the comF gene

Validation