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.

- 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

TopSpin 3.6.2 (Bruker) for all NMR experiments

Data analysis

TopSpin 3.6.2 (Bruker) for all analysis of NMR spectra

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

Source data are available for Figs. 2–5 and Supplementary Figs. 2, and 4–8. Source data are provided with this paper.
Human research participants

Policy information about studies involving human research participants and Sex and Gender in Research.

Reporting on sex and gender  Not applicable
Population characteristics Not applicable
Recruitment Not applicable
Ethics oversight Not applicable

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 means and error bars of the kex values were obtained from 50 data sets of intensities constructed by Monte Carlo simulation based on the standard deviation of the noise signal for the region of 1D 1H spectrum in which no signal is present. The 50 data sets were sufficient sample size to hold our claim that kopen(in-cell) is higher than kopen(in vitro).

Data exclusions
No data was excluded from the analysis.

Replication
Replicates are indicated in Figure legends. The in-cell water magnetization transfer experiments for hairpin RNA, hprNA14, has been repeated three times and at each time, the similar kex values were obtained. The in-cell measurements are reproducible enough to hold our claim that kopen(in-cell) is higher than kopen(in vitro).

Randomization
Randomization method was not utilized. This is not a clinical trial (i.e., no animals or humans were involved) and different strains were not being compared to each other.

Blinding
Blinding is not relevant for this study because no group allocation was performed.

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

Eukaryotic cell lines
Policy information about cell lines and Sex and Gender in Research

Cell line source(s)
HeLa cells were purchased from Riken BRC Cell Bank (Ibaraki, Japan).

Methods
n/a Involved in the study
☒ ChIP-seq
☒ Flow cytometry
☒ MRI-based neuroimaging
| Authentication | Cell lines were not authenticated after they were received. |
|----------------|------------------------------------------------------------|
| Mycoplasma contamination | The cell lines were not tested for mycoplasma contamination. |
| Commonly misidentified lines (See ICLAC register) | No commonly misidentified cell lines were used in this study. |