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corresponding author(s):	Yujie Sun
Last updated by author(s):	Shipeng Shao

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 our Editorial Policies and the Editorial Policy Checklist.

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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 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)	nt
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 Give <i>P</i> 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 <i>d</i> , Pearson's <i>r</i>), indicating how they were calculated	
Our web collection on <u>statistics for biologists</u> contains articles on many of the points above.	
Software and code	

Software and code

Policy information about <u>availability of computer code</u>

Data collection

All data are collected on a custom Olympus IX83 inverted microscope equipped with a 100X UPlanSApo, N.A. = 1.49, oil-immersion phase objective and Andor iXon Ultra EMCCD.

RStudio, MatLab and Microsoft Excel.

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 Research 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 list of figures that have associated raw data
- A description of any restrictions on data availability

All data generated and analyzed during the current study are available from the corresponding author on reasonable request.

Life sciences study design

Commonly misidentified lines (See <u>ICLAC</u> register)

No misidentified cell lines

Life deletiees	s study design			
All studies must disclose o	n these points even when the disclosure is negative.			
Sample size more t	more than 10 cells were used to extract the single molecule dynamics			
Data exclusions no data	no data were excluded in the samples			
Replication Each ex	Each experiment was repeated 2-3 times			
Randomization n/a	(n/a			
Blinding n/a	n/a			
	or specific materials, systems and methods			
•	authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, evant 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 & experime	ental systems Methods			
n/a Involved in the study Antibodies Eukaryotic cell lines Palaeontology and Animals and other of Human research pa Clinical data Dual use research of	ChIP-seq Chip-seq Flow cytometry archaeology MRI-based neuroimaging organisms rticipants			
Antibodies Antibodies used	c-Fos (9F6) Rabbit m&b CST#2250, c-lun (60A8) Rabbit m&b CST#9165, Flag (D6W5R) Rabbit m&b CST#14793, GAPDH Mouse			
, and bodies doed	c-Fos (9F6) Rabbit mAb CST#2250, c-Jun (60A8) Rabbit mAb CST#9165, Flag (D6W5B) Rabbit mAb CST#14793, GAPDH Mouse Monoclonal (Proteintech-60004-1-lg)			
Validation	Validation of the all the antibodies we used were described in the manufacturers website			
Eukaryotic cell lir	es			
Policy information about <u>c</u>	ell lines			
Cell line source(s)	MDA-MB-231 was a gift from Dr. Wei Guo (Department of Biology, University of Pennsylvania)			
Authentication	None of the cell lines were authenticated within 1 year			
Mycoplasma contamination	ntamination All cell lines were tested negative for mycoplasma contamination			