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Corresponding author(s): Bustamante, Carlos

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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, see our <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

Statistics

For	For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.						
n/a	Cor	Confirmed					
	\square	The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement					
	\boxtimes	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly					
\boxtimes		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.					
\boxtimes		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 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)					
\boxtimes		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.					
\boxtimes		For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings					
\boxtimes		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					
		Our web collection on statistics for biologists contains articles on many of the points above.					

Software and code

Policy information a	about <u>availability of computer code</u>
Data collection	The optical tweezers instrument was run using custom code in LabVIEW 8.1.
Data analysis	The analysis was performed using custom code in MATLAB R2016a.

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 <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 data that support the findings of this study are available from the corresponding author upon reasonable request.

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 <u>nature.com/documents/nr-reporting-summary-flat.pdf</u>

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Sample size is number of single-molecule trajectories for each test group. At least 50 traces (~10000 bursts) was taken for each group.				
Data exclusions	Some data for the PWD for sake of signal-to-noise ratio, but analyses including all of the data are also included.				
Replication	The single-molecule trajectories are independent of each other. The experiment has been repeated succesfully on differing days, too.				
Randomization	Each experiment in the same group is the same, so there is no need for randomization.				
Blinding	Experimental groups were not blinded, but the recording of data is hands-off and the analysis methods were identical across groups.				

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			Methods		
n/a	n/a Involved in the study		Involved in the study		
	Antibodies	\boxtimes	ChIP-seq		
\boxtimes	Eukaryotic cell lines	\boxtimes	Flow cytometry		
\boxtimes	Palaeontology and archaeology	\boxtimes	MRI-based neuroimaging		
\boxtimes	Animals and other organisms				
\boxtimes	Human research participants				
\boxtimes	Clinical data				
\boxtimes	Dual use research of concern				

Antibodies

Antibodies used phi29 gp8 (major capsid protein): rabbit (Rockland, #X811), digoxigenin For anti-gp8 (anti-dig): pulldown assay: Crosslink to bead, test pulldown of capsid (dig-DNA) in PAGE (AGE) Validation