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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 <u>Authors & Referees</u> and the <u>Editorial Policy Checklist</u>.

Statistical parameters

	en statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. figure legend, table legend, main c, or Methods section).
n/a	Confirmed
	The <u>exact sample size</u> (n) for each experimental group/condition, given as a discrete number and unit of measurement
	An indication of 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.
\boxtimes	A description of all covariates tested
X	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
\boxtimes	A full description of the statistics including <u>central tendency</u> (e.g. means) or other basic estimates (e.g. regression coefficient) AND <u>variation</u> (e.g. standard deviation) or associated <u>estimates of uncertainty</u> (e.g. confidence intervals)
\boxtimes	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 <i>Give P values as exact values whenever suitable.</i>
\times	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
	Clearly defined error hars

Our web collection on <u>statistics for biologists</u> may be useful.

Software and code

Policy information about availability of computer code

Data collection We used NIS Elements (version 3.22.15) to collect imaging data.

Data analysis We used custom MATLAB (R2018a) code to quantify fluorescence intensity. We also used custom Python code to analyze RNA-sequencing data. We also used BowTie 2 and custom Perl scripts to analyze RNA-sequencing data. The code will be available upon

request after publication.

State explicitly what error bars represent (e.g. SD, SE, CI)

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 upon request. 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 <u>availability of data</u>

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

Figure 1 and supplemental tables S1 and S2 have associated raw sequencing data. Most of the figures have associated raw images. All of this raw data will be available upon request after publication.		
Field-spe	ecific reporting	
Please select the b	est 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 t	the document with all sections, see <u>nature.com/authors/policies/ReportingSummary-flat.pdf</u>	
Life scier	nces study design	
All studies must dis	sclose on these points even when the disclosure is negative.	
Sample size	For quantification of individual cells, we measured 30 or more cells. We considered a given experiment that included at least 30 cells to be one experimental replicate, and calculated the standard deviation of data from at least three experiments. This sample size was sufficient to support our conclusions as it led to standard deviations across replicates that were much smaller than the experimental effects from which were drawing conclusions.	
Data exclusions	No data was excluded.	
Replication	All quantified experiments were performed independently at least three times. There were no experiments that could not be replicated or reproduced.	
Randomization	Randomization was not relevant to this study, as all experiments are performed on bacterial strains grown in broth culture, which were then split in different treatment conditions.	
Blinding	No blinding was used. Blinding was not relevant to this study, as we used the same set of data acquisition and analysis parameters for each biological sample.	
Reportin	g for specific materials, systems and methods	
	<u> </u>	
Materials & experimental systems Methods		
n/a Involved in the study		
	ological materials ChIP-seq	

Unique biological materials

Animals and other organisms Human research participants

Policy information about <u>availability of materials</u>

Antibodies

Palaeontology

Eukaryotic cell lines

Obtaining unique materials All strains and plasmids will be available upon request after publication.

Flow cytometry

MRI-based neuroimaging