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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	all st	atistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.		
n/a	a Confirmed			
	\boxtimes	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 <u>statistics for biologists</u> contains articles on many of the points above.		

Software and code

Policy information	about <u>availability of computer code</u>
Data collection	No software was used.
Data analysis	NIS Elements 4.5 Advance Research software (Nikon), Image J

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

This article presents representative data that support the presented approach. Additional data are available in the supporting research papers (gel electrophoresis, digital immunoassays, and immunocytochemistry). Primary data underlying the figures shown in this protocol are available upon a reasonable request from the corresponding authors.

Field-specific reporting

Life sciences

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.

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 dis	close on these points even when the disclosure is negative.
Sample size	Typically 3 samples were used for each experiment.
Data exclusions	No data were excluded from the analysis
Replication	All attempts of replication were successful.
Randomization	Allocation was not necessary as we used negative controls in our experiments.
Blinding	Blinding was not relevant for our studies.

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

Antibodies

Antibodies used	Polyclonal horse anti-mouse antibody (Vector Laboratories); monoclonal mouse anti-PSA antibody (ab403, Abcam); biotinylated anti- PSA antibody (BAF1344, R&D Systems); rabbit anti-HER2 antibody (ab134182, Abcam); biotinylated anti-rabbit antibody (111-065-144, Jackson ImmunoResearch)
Validation	Negative controls were performed to show specific binding.

Eukaryotic cell lines

Policy information about <u>cell lines</u>	
Cell line source(s)	Breast cancer cell lines BT-474 (ATCC HTB-20), MCF-7 (ATCC HTB-22), and MDA-MB-231 (ATCC HTB-26).
Authentication	Describe the authentication procedures for each cell line used OR declare that none of the cell lines used were authenticated.
Mycoplasma contamination	Confirm that all cell lines tested negative for mycoplasma contamination OR describe the results of the testing for mycoplasma contamination OR declare that the cell lines were not tested for mycoplasma contamination.
Commonly misidentified lines (See <u>ICLAC</u> register)	Name any commonly misidentified cell lines used in the study and provide a rationale for their use.