nature portfolio

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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 <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

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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)
	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>
x	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 statistics for biologists contains articles on many of the points above

Software and code

Policy information about availability of computer code

Data collection

 $HPLC: Shimadzu\ Corporation\ Lab Solutions\ version\ 5.86;$

LC-MS/MS: Mascot version 2.3.02 (Matrix Science, London, UK);

Western blotting imaging: Tanon MP version 1.0 (Tanon, Shanghai, China); Integrated density of the Western blotting bands: ImageJ software (1.53K);

Glucose: SBA-40D biosensor analyzer (Institute of Biology, Shandong Academy of Sciences, Jinan, China);

OD600: MAPADA V-1100D spectrophotometer (MAPADA Instruments, Shanghai, China);

Molecular weight: on-line tool (https://web.expasy.org/compute_pi/);

Data analysis

Excel 2019 (Microsoft): mean, SE, P value, plotting;

ProteoWizard (3.0.10577): MS/MS data processing;

Mascot version 2.3.02 (Matrix Science, London, UK): protein identification;

ChemDraw 2014: chemical structure; Office 2021 v2409 (Microsoft): plotting;

Origin 8.5 (OriginLab): plotting;

SPSS Statistics 19.0 (IBMA): one-way ANOVA;

R language version 4.3.3, R Studio 2023.12.1 Build 402: plotting, curve fitting;

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

 $Data \ supporting \ the \ findings \ of \ this \ study \ are \ available \ within \ the \ paper \ and \ its \ Supplementary \ Information \ file. \ The \ GenBank \ (https://www.ncbi.nlm.nih.gov/loop) \ and \ are \ available \ within \ the \ paper \ and \ its \ Supplementary \ Information \ file.$ genbank/) accession numbers and codon-optimized nucleotide sequences of the genes referenced in this study are provided in this paper. The mass spectrometry proteomics data have been deposited to the ProteomeXchange Consortium via the PRIDE partner repository with the dataset identifier PXD056539 (https:// www.ebi.ac.uk/pride/archive/projects/PXD056539). Source data are provided with this paper.

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	No human participants involved.
Reporting on race, ethnicity, or other socially relevant groupings	No human participants involved.
Population characteristics	No human participants involved.
Recruitment	No human participants involved.
Ethics oversight	No human participants involved.

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 belo	ow that is the best fit for your research	If you are not sure, read the appropriate sections before making your selection. $ \\$
x 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 No specific calculations were used to determine sample size for each experiment. This experiment was performed on bacteria fermentation

> and transformation for the production of chemicals, involving a large population of individual cells. All presented data represent measurements from at least 3 biological replicates, grown and performed independently. To ensure the reproducibility, some experiments were performed for several times with relevant independent replicates. For each experiment, the detailed sample sizes were stated in legend. For kinetic analysis, 3 biological replicates per substrate concentration group were performed.

Data exclusions No data were excluded from our analyses.

Blinding

Replication All the fermentations, biotransformations and enzymatic assays were performed at least three times. The exact number of independent experiments were described in the legend.

Randomization At least three single colonies of each recombinant strain were randomly selected from plates, each of which was subjected to independent

flask cultures, batch fermentations and chemical analyses.

Blinding was not relevant to this study. We designed and carried out the experiments to achieve our goal. All samples were extracted and assayed in the same way, without any differences.

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 & experime	ntal sys	tems Methods
n/a Involved in the study		n/a Involved in the study
Antibodies		ChIP-seq
Eukaryotic cell lines		Flow cytometry
Palaeontology and a	ırchaeolog	y MRI-based neuroimaging
Animals and other o	rganisms	
Clinical data		
Dual use research of	fconcern	
Plants		
Antibodies		
Antibodies used	antibody	tern blotting analysis, anti-Flag tag mouse monoclonal antibody (Cat. No. D191041), anti-6×His tag mouse monoclonal (Cat. No. D191001), and horseradish peroxidase-conjugated goat anti-mouse IgG (Cat. No. D110087) from Sangon Biotech ii, China) were used at 1:4000, 1:4000, and 1:5000, respectively.
Validation	The webs	antibodies used in this study are commercially available and validation statements are given on manufacturer's websites. site of Cat. No. D191041, Cat. No. D191001, and Cat. No. D110087 is https://store.sangon.com/productDetail? nfo.code=D191041, https://store.sangon.com/productDetail?productInfo.code=D191001 and https://store.sangon.com/petail?productInfo.code=D110087, respectively.
Eukaryotic cell line	es	
Policy information about <u>ce</u>	ell lines ar	nd Sex and Gender in Research
Cell line source(s)	(V	lo eukaryotic cell lines involved.
Authentication	(V	lo eukaryotic cell lines involved.
Mycoplasma contamination	V	lo eukaryotic cell lines involved.
Commonly misidentified I (See <u>ICLAC</u> register)	ed lines No eukaryotic cell lines involved.	
Palaeontology and	d Arch	naeology
Specimen provenance	This stud	y did not involve specimens.
Specimen deposition	This study did not involve specimens.	
Dating methods	This study did not involve specimens.	
Tick this box to confirm	m that th	e raw and calibrated dates are available in the paper or in Supplementary Information.
Ethics oversight	This study did not involve specimens.	
Note that full information on the	he approva	al of the study protocol must also be provided in the manuscript.
Animals and othe	r rese	arch organisms
Policy information about str Research	udies inv	olving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in
Laboratory animals	This stud	y did not involve animals.
Wild animals	This stud	y did not involve animals.
Reporting on sex	This stud	y did not involve animals.
Field-collected samples	This stud	y did not involve animals.
Ethics oversight	This stud	y did not involve animals.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

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Policy information about <u>clinical studies</u>				
All manuscripts should comply with the ICMJEguidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.				
Clinical trial registration	This study did not involve clinical trial.			
Study protocol	This study did not involve clinical trial.			
Data collection	This study did not involve clinical trial.			
Outcomes	This study did not involve clinical trial.			

Dual use research of concern

Policy information about <u>dual use research of concern</u>

Hazards

Could the accidental, deliberate or reckless misuse of agents or technologies generated in the work, or the application of information presented in the manuscript, pose a threat to:

No	Yes	i e
×		Public health
x		National security
x		Crops and/or livestock
x		Ecosystems
x		Any other significant area

Experiments of concern

Does the work involve any of these experiments of concern:

No	Yes
x	Demonstrate how to render a vaccine ineffective
x	Confer resistance to therapeutically useful antibiotics or antiviral agents
x	Enhance the virulence of a pathogen or render a nonpathogen virulent
x	Increase transmissibility of a pathogen
x	Alter the host range of a pathogen
×	Enable evasion of diagnostic/detection modalities
×	Enable the weaponization of a biological agent or toxin
x	Any other potentially harmful combination of experiments and agents

Plants

Seed stocks	This study did not involve plants.	
Novel plant genotypes	This study did not involve plants.	
Authentication	This study did not involve plants.	

ChIP-seq			
Data deposition			
Confirm that both rav	w and fi	nal processed data have been deposited in a public database such as GEO.	
Confirm that you hav	e depos	ited or provided access to graph files (e.g. BED files) for the called peaks.	
Data access links May remain private before publ	ication.	This study did not involve ChIP-seq experiment.	
Files in database submission	n	This study did not involve ChIP-seq experiment.	
Genome browser session (e.g. <u>UCSC</u>)	ı	This study did not involve ChIP-seq experiment.	
Methodology			
Replicates	This stu	udy did not involve ChIP-seq experiment.	
Sequencing depth	This stu	udy did not involve ChIP-seq experiment.	
Antibodies	This stu	udy did not involve ChIP-seq experiment.	
Peak calling parameters	This stu	udy did not involve ChIP-seq experiment.	
Data quality	This stu	udy did not involve ChIP-seq experiment.	
Software	This stu	udy did not involve ChIP-seq experiment.	
Flow Cytometry			
Plots			
Confirm that:	Confirm that:		
The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).			
The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).			
All plots are contour plots with outliers or pseudocolor plots.			
A numerical value for	numbe	r of cells or percentage (with statistics) is provided.	
Methodology			
Sample preparation		This study did not involve flow cytometry.	
Instrument		This study did not involve flow cytometry.	
Software		This study did not involve flow cytometry.	
Cell population abundance		his study did not involve flow cytometry.	
Gating strategy		This study did not involve flow cytometry.	
Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.			
Magnetic resonance imaging			
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Experimental design			
Design type		This study did not involve magnetic resonance imaging.	
	Design specifications This study did not involve magnetic resonance imaging.		
Behavioral performance measures This study did not involve		This study did not involve magnetic resonance imaging.	

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Acquisition				
Imaging type(s)	This study did not involve magnetic reso	nance imaging.		
Field strength	This study did not involve magnetic reso	nance imaging.		
Sequence & imaging parameters	This study did not involve magnetic reso	nance imaging.		
Area of acquisition	This study did not involve magnetic reso	nance imaging.		
Diffusion MRI Used	Not used			
Preprocessing				
Preprocessing software This study did not involve magnetic resonance imaging.				
Normalization	This study did not involve magnetic resonance imaging.			
Normalization template	This study did not involve magnetic resonance imaging.			
Noise and artifact removal	This study did not involve magnetic resonance imaging.			
Volume censoring	This study did not involve magnetic resonance imaging.			
Statistical modeling & infere	ee			
Model type and settings	nis study did not involve magnetic resonance	: imaging.		
Effect(s) tested	This study did not involve magnetic resonance imaging.			
Specify type of analysis: W	le brain ROI-based Botl	١		
Statistic type for inference	This study did not involve magnetic resonance imaging.			
(See Eklund et al. 2016)				
Correction	nis study did not involve magnetic resonance	e imaging.		
Models & analysis				
n/a Involved in the study				
Functional and/or effective connectivity				
Graph analysis				
Multivariate modeling or p	lictive analysis			
Functional and/or effective connection	This study did not involve ma	gnetic resonance imaging.		
Graph analysis	This study did not involve ma	gnetic resonance imaging.		

This study did not involve magnetic resonance imaging.

Multivariate modeling and predictive analysis