natureresearch

Corresponding author(s): Harry J. Gilbert

Revised version

Initial submission

Final submission

Life Sciences Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form is intended for publication with all accepted life science papers and provides structure for consistency and transparency in reporting. Every life science submission will use this form; some list items might not apply to an individual manuscript, but all fields must be completed for clarity.

For further information on the points included in this form, see Reporting Life Sciences Research. For further information on Nature Research policies, including our data availability policy, see Authors & Referees and the Editorial Policy Checklist.

Experimental design

1.	Sample size			
	Describe how sample size was determined.	All the enzyme kinetic data in Supplementary Tables 2 and 3 and reaction profiles shown in Fig. 2 represent a minimum of technical triplicates. The data for a few of the enzymes were biological triplicates. The microbial experiments reported in Supplementary Table 4, Fig. 5bc, Fig. 6 and Supplementary Fig. 2 represent 3 biological replicates with each replicate repeated twice. The whole cell assays reported in Fig. 5e are biological triplicates. Analysis of microbial products are all biological replicates with n = 2 in Figure 2 and 4, Supplementary Figures 7 and 8 and n = 4 in Supplementary Figure 12. Localization experiments using antibodies in Fig. 5a and whole cell assays in Fig. 5e are examples of biological replicates n = 3. The number of biological replicates in the RT-PCR data shown in Supplementary Fig. 1b and 1c are n=6 and n=3, respectively. The error bars where shown were standard errors of the mean. The proteomics described in Figure 5f, Supplementary Figure 10 and Supplementary table 5 are examples of biological replicates with n = 2.		
2.	Data exclusions			
	Describe any data exclusions.	No data were excluded from the analysis.		
3.	Replication			
	Describe whether the experimental findings were reliably reproduced.	All attempts at replication were successful.		
4.	Randomization			
	Describe how samples/organisms/participants were allocated into experimental groups.	The randomization was not relevant in this study where only in intro assay were performed.		
5.	Blinding			
	Describe whether the investigators were blinded to group allocation during data collection and/or analysis.	The blinding was not relevant in this study where only in intro assay were performed.		

Note: all studies involving animals and/or human research participants must disclose whether blinding and randomization were used.

6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

n/a	Con	firmed
	\square	The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)
	\boxtimes	A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
	\square	A statement indicating how many times each experiment was replicated
\boxtimes		The statistical test(s) used and whether they are one- or two-sided (note: only common tests should be described solely by name; more complex techniques should be described in the Methods section)
\boxtimes		A description of any assumptions or corrections, such as an adjustment for multiple comparisons
\boxtimes		The test results (e.g. P values) given as exact values whenever possible and with confidence intervals noted
	\square	A clear description of statistics including <u>central tendency</u> (e.g. median, mean) and <u>variation</u> (e.g. standard deviation, interquartile range)
	\square	Clearly defined error bars
		See the web collection on statistics for biologists for further resources and guidance.

Software

Policy information about availability of computer code

7. Software

Describe the software used to analyze the data in this study.

Growth curves and Reat time-qPCR data were analyzed using Prism Graphpad software or Gen5 software (BioTek). NMR data was analyzed with Azara suite of programs (v. 2.8, copyright 1993-2017, Wayne Boucher and Department of Biochemistry, University of Cambridge, unpublished) and chemical shift assignment was performed using CCPNMR Analysis v2.4.

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). *Nature Methods* guidance for providing algorithms and software for publication provides further information on this topic.

Materials and reagents

Policy information about availability of materials

```
8. Materials availability
```

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a for-profit company.

9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).

10. Eukaryotic cell lines

- a. State the source of each eukaryotic cell line used.
- b. Describe the method of cell line authentication used.
- c. Report whether the cell lines were tested for mycoplasma contamination.
- d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by ICLAC, provide a scientific rationale for their use.

All unique materials used are readily available from the authors or from standard commercial sources.

Protein cell localization was performed by immunochemical detection using primary rabbit polyclonal antibodies (Eurogentec) generated against the proteins BT0264 and BT4662 and secondary goat anti-rabbit antibodies (sc-2004, Santa Cruz Biotechnology). Primary rabbit polyclonal antibodies specificity was validated by the specific immunochemical detection of purified protein.

No eukaryotic cell lines were used.

Animals and human research participants

Policy information about studies involving animals; when reporting animal research, follow the ARRIVE guidelines

11. Description of research animals

Provide details on animals and/or animal-derived materials used in the study.

No animals were used.

Policy information about studies involving human research participants

12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.

The study did not involve human research participants.