

## Supplementary Information

Table S1. Bacterial strains and plasmids used in this study.

Strains or plasmids	Relevant characteristics <sup>a</sup>	Source or reference
<b>Strains</b>		
<i>E. coli</i>		
DH5 $\alpha$	F <sup>-</sup> <i>endA1 glnV44 thi-1 recA1 relA1 gyrA96 deoR nupG purB20 <math>\phi</math>80dlacZ<math>\Delta</math>M15 <math>\Delta</math>(lacZYA-argF)U169, hsdR17(<i>r</i>K<sup>-</sup><i>m</i>K<sup>+</sup>), <math>\lambda</math><sup>-</sup></i>	Lab collection
DH-26	DH5 $\alpha$ pLPRS-RFP	This study
DH-31	DH5 $\alpha$ pLPRS-tetA-RFP	This study
<i>C. glutamicum</i>		
ATCC 13032	Wild-type strain	Lab collection
QW36	pEC-A263-RFP, derived from ATCC 13032	This study
QW37	pEC-R152-RFP, derived from ATCC 13032	This study
QW45	<i>lysC</i> (Q298G), derived from ATCC 13032	This study
QW48	LPRS-A263- <i>lysC</i> , derived from QW45	This study
QW53	LPRS-R152-hom, derived from QW45	This study
QW54	LPRS-R357-hom, derived from QW45	This study
QW55	LPRS-A263- <i>lysC</i> , LPRS-R357-hom, derived from QW45	This study
<b>Plasmids</b>		
pAmp-Ori	Cloning vector	Lab collection
pJ-RFP	Expression vector	Lab collection
pK18mobsacB	Kan <sup>R</sup> , SacB	Lab collection
pEC-XK99E	Expression vector	Lab collection
pLPRS-RFP	BBa_J23100, <i>Lb. plantarum</i> lysine riboswitch (LPRS), RFP	This study
pG14A-RFP	LPRS (G14A), derived from pLPRS-RFP	This study
pLPRS-tetA-RFP	tetA, derived from pLPRS-RFP	This study
pEC-A263-RFP	LPRS-A263 fused with <i>RFP</i> gene, derived from pEC-XK99E	This study
pEC-R152-RFP	LPRS-R152 fused with <i>RFP</i> gene, derived from pEC-XK99E	This study
pA263- <i>lysC</i>	LPRS-A263 fused with <i>lysC</i> gene, derived from pK18mobsacB	This study
pR152-hom	LPRS-R152 fused with <i>hom</i> gene, derived from pK18mobsacB	This study
pR357-hom	LPRS-R357 fused with <i>hom</i> gene, derived from pK18mobsacB	This study

Table S2. Primers used in this study.

Primer	Sequence	Description
LPRS-F	atcgaaagaagaggatgcg	For <i>Lb. plantarum</i> lysine
LPRS-R	ctgccattgcagaaacctcc	riboswitch amplification
RFP-F	atggcttctccgaagac	For <i>RFP</i> amplification
RFP-R	ttaagcaccgggtggagtgc	
BR-F	agtagctagcactgtacctag	For backbone of pJ-RFP
BR-R	gatgctgaaaagaacgccg	amplification
tetA-F	atgcaagtcgacctgctgg	For <i>tetA</i> amplification
tetA-R	cctctccagatcctcctccagatccacctccaagggtcgag gtggccccggc	
RSlib-F	NNNNNNNNNNNNNNNNNNNNgaaatcat gctgaaaagaacg	For pLPRS-tetA-RFP amplification
RSlib-R	NNNNNNNNNNNNNNNNNNNNcctcttcttt cgatataactcaac	
lysC-F	aggtggccctggctgtacag	For <i>lysC</i> amplification
lysC-R	cttttagcgtccggtgcctgc	
hom-F	atcatgacctcagcatctgc	For <i>hom</i> amplification
hom-R	attagtcctttcgaggcgg	
K18-F	GGATCCCCTGACACGAGGTAGTTatgacc tcagcatctgccccaaag	For backbone of pK18mobsacB amplification
K18-R	GGCCTGCGCTAGCGCAGGTAGTACctatt actttgttcggccaccc	

Table S3. Predicted results of lysine riboswitch in *C. glutamicum* ATCC 13032 using Riboswitch Scanner.

Genome_ID	Genome_Name	Taxonomy_name		Riboswitch Location	Riboswitch Strand	Riboswitch class	Downstream gene location
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	66279-66442	-	FMN	65504-66197
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	554235-554393	-	cobalamin	552944-554129
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	741074-741181	-	TPP	740224-741016
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	868388-868556	-	ydaO-yuaA	867799-868381
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	1126306-1126415	+	TPP	1127012-1128353
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	1371634-1371746	-	TPP	1369550-1369877
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	1372540-1372656	+	SAM-IV	1372804-1373134
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	1542916-1543023	-	TPP	1539816-1542108
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	2150221-2150334	+	TPP	2150334-2151000
NC_003450.3	Corynebacterium glutamicum ATCC 13032	Actinobacteria	201174	2320608-2320796	-	ydaO-yuaA	2319965-2320595

Table S4. Predicted results of lysine riboswitch in *Lb. plantarum* WCFS1 13032 using Riboswitch Scanner.

Genome_ID	Genome_Name	Taxonomy_name		Riboswitch Location	Riboswitch Strand	Riboswitch class	Downstream gene location
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	99249-99354	+	TPP	99432-100224
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	202607-202700	-	TPP	201934-202504
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	482228-482273	+	PreQ1	482281-482788
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	761448-761599	+	glmS	761631-763449
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	928916-929097	+	lysine	929240-930692
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	1189828-1189933	+	SMK_box	1191254-1192757
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	1705901-1706022	-	FMN	1705192-1705777
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	1741167-1741232	-	fluoride	1739825-1741115

NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	1874486- 1874649	-	ykoK	1872642- 1874385
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	2410476- 2410574	+	purine	2410633- 2411974
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	2519515- 2519559	-	PreQ1	2518513- 2519494
NC_004567.2	Lactobacillus plantarum WCFS1	Firmicutes	1239	2968731- 2968829	-	purine	2966961- 2968653

Table S5. The sequences of the engineered *Lb. plantarum* lysine riboswitches.

Engineered LPRS clone	Sequence
Lysine-activated	
4	cccgtgagacggctgtagcacc
17	gtgagcgaacatatgatacatcgg
29	gtacaccgcacaccagcagcttg
53	ggtgtgacacgggtcacc
124	gaggccggcacagagtgtctcg
171	tcgcgtctggtgattgatgtctctg
192	gaccggcgtaagctcgagatagt
209	cgggtgagcgggtgtatatgcaag
263	acgatacgtcgcgcagtgtgga
284	gctggaccgctccatctgcggac
Lysine-repressed	
31	cgtcaggtctccgatgagtcg
46	agggctgtactcgattcgg
61	tctacgtgaagcattctggaagatg
84	gaccacatgcgatgactcagcag
139	ctcccgaatgattgcaatagg
152	tgactggtctatcaggctgtacg
217	ccgcatcgccgtaggtggcag
298	catcagccagttggcttgcgt
314	aggcacgcaccgttagcgcgcg
352	tccagtgcagtgttgagatatgtc