

Supplementary material

Table S1 Intracellular metabolic profiles obtained by MSFTA-GCMS analysis of cell pellets. Bacterial cultures were sampled at OD₆₀₀ 1.5, 2.5, 4.5 and 6.5. A methanol-chloroform extraction was used to isolate metabolites prior to being derivatized with MSTFA and analysed by GCMS. Data is ordered by p-value at OD₆₀₀ 4.5. Asterisks indicate significant differences (* $P \leq 0.05$, ** $P \leq 0.01$, two-tailed Student's *t* test). Multiple MSFTA derivatives of the same metabolite are indicated by Arabic numerals in brackets. ND = not detected.

Metabolite	Fold change (<i>lasI rhII</i> mutant / wild-type)			
	OD ₆₀₀ = 1.5	OD ₆₀₀ = 2.5	OD ₆₀₀ = 4.5	OD ₆₀₀ = 6.5
1-Glycerophosphoric acid	1.3	2.1	27**	3**
2-Hydroxypentanedioic acid	1.2	1.9	5.4**	3.1**
3-Pyridinecarboxamide (Nicotinamide)	1.1	1.2	2.2**	2.8**
Pentose-phosphate	1.2	1.1	16.5**	10.1**
Phenylalanine (2)	0.8	0.6	0.5**	2.3
3-Hydroxydecanoic acid	0.9	1.1	0.3**	0.1**
Trehalose	1.3	1.1	4.3**	ND
Hypoxanthine (6- Hydroxypurine)	1.2	1.7	2.4**	2.5
Glutaric acid	1.2	0.7	0.4**	1.2
Monosacharide	1.1	1.1	1.8**	1.9**
Aromatic	0.9	1.1	2.8**	4.4*
Citric acid	0.9	1.6	2**	2.7**
Fructose 6-phosphate	1.1	1.8	2.8**	2.9*
5-Aminopentanoic acid	1.1	1	3.5**	6.5
Myo-inositol	1	1.1	1.6**	1.6**
Citramalic acid	0.8	0.8	1.8**	3.8**
Ribose	1.2	1.7	1.9**	2.5**
Methionine	1.2	0.7	0.7**	0.5**
Ribofuranose	1.2	1.1	0.6**	0.7*
Malic acid	0.8	1.1	1.6**	1.3
Hexose-phosphate	0.8	1.8	3.1**	2.4*
Aspartic acid	1.2	0.8	0.6**	0.8
Tyrosine (1)	1.3	0.9	0.6*	0.8
Adenine	1.1	0.8	0.6*	0.7*
Hexose-phosphate	0.9	1.8	2.7*	2*
Glutamic acid	0.7	0.7	2*	2.5
Pyrimidine	1.5	1.2	1.7*	1.9
Alkane	1.2	0.7	0.6*	0.5**
Putrescine (2)	0.8	1.2	4.3*	5.9*
Phenylalanine (1)	1	0.9	0.5*	1.9
Glucose or galactose	1.8	1.1	0.7*	0.5
Amino acid	1	0.7	1.4*	1.2
Alkane	1	0.8	0.5*	0.5*

Long chain alcohol	1.4	0.8	0.5*	0.6**
Hexadecanoic acid	1.2	0.7	0.5*	0.5**
N-acetylglutamic acid (1)	0.9	1.2	1.9*	1.9
Mono-unsaturated fatty acid (1)	1	1	0.5*	0.5*
Hexose-phosphate	1.2	3.2	2.9*	4.3**
Tyrosine (2)	1	0.7	0.5*	1.9
Alkane	1	0.7	0.6	0.4*
Alkane	1.4	0.6	0.5	ND
Aromatic	0.8	1	0.7	0.7
Octadecanol	1.3	0.5	0.5	0.6
Nonanoic acid	1.2	0.8	0.5	0.7*
d-altro-2-Heptulose, 7- (dihydrogen phosphate)	1.2	3.8	3	5.2**
Octadecanoic acid	1.3	0.7	0.5	0.4**
Spermidine	1	1.2	1.7	3.5
Polyamine	1	1.3	2.7	4.8
5'-Adenylic acid (AMP)	1.5	0.8	0.3	0.1*
Putrescine (1)	0.6	1.3	6	5.3
Lysine	0.9	0.6	1.6	3.3
1-Monopalmitoylglycerol	1.4	1.1	0.6	0.7*
Cysteine	0.6	0.5	0.7	0.6
Aromatic	1.5	0.7	0.5	0.3**
Succinate	1.1	1	1.2	1.4*
Octacosane	1.3	0.7	0.5	0.5*
Glycerol	1	0.9	1.2	1.2
Arginine	1	0.8	1.4	2.3**
Alkane	1	0.6	0.5	0.5**
Long chain alcohol	1.3	1	1.5	1.1
Fumaric acid	1.2	1.1	1.2	1.4**
Adenosine	1	0.9	0.7	0.1
Threonine (2)	1.1	1	0.2	1.1
N-acetyl glucosamine	1.1	1	1.2	1
Alkane	1.4	0.6	0.6	0.5
Pyroglutamic acid	1.1	1.1	1.2	0.9
5'-Uridylic acid (UMP)	1.9	1.4	1.8	3.2
Glucose or Galactose	1.1	1.2	0.9	1.4*
Pantothenic acid	0.9	1.1	1.3	1.5*
Valine	1.1	1.1	1.2	2.3
Threonine (1)	1.3*	1	0.2	0.8
Glucose or Galactose	1.2	1.3	0.9	2**
Glycine	0.9	1.1	1.2	1
Proline	1.1	0.8	1.1	0.9
Beta-alanine	1	1.1	1.2	0.4*
Alkane	1.3	0.6	0.7	ND
Ornithine	0.8	0.5	0.6	4*

Monosacharide	0.9	0.9	0.9	ND
Tetradecanoic acid	1	0.7	0.8	1
Glucose or Galactose	2.2	1.4	0.9	1.4*
4-Aminobutyric acid	1.1	1	0.9	0.5
Ribofuranose	1.7	1.3	1.3	0.6
Uracil	1.3	1.2	1	1.1
<i>S</i> -Methyl-5-thioadenosine	1.5	1.5	0.9	2.1
N-acetyl glutamic acid (2)	1.1	1.1	0.9	0.7
Ethanol, 2-amino-	0.7	0.5	0.9	4.1
2-Glycerophosphoric acid	1	1	1	1.1
Xanthine	1	1.3	0.9	6.9
Ethanedithioic acid <i>S</i> -(3-methylbutyl) ester	1	0.8	1	1.3
2-Methylbenzoic acid (<i>o</i> -Toluic acid)	0.8	0.9	1	1.8

Table S2 Amino acid and polyamine metabolite profiles as determined by LCMS. Student's *t*-tests were calculated relative to the wild-type (WT, for the *lasI rhII* mutant) or relative to the *lasI rhII* mutant (LIRI, for chemical complementation(s)). Complementation values are shown where significant ($p \leq 0.05$). Asterisks indicate significant differences (* $P \leq 0.05$, ** $P \leq 0.01$, two-tailed Student's *t* test). "NS" indicates no significant change relative to the non-complemented culture.

Metabolite	OD1.5					OD4.5				
	WT	LIRI	BHL	OdDHL	BHL + OdDHL	WT	LIRI	BHL	OdDHL	BHL + OdDHL
1-methyl histidine	0.5	0.1	NS	NS	NS	0.2	0.3	NS	NS	NS
3-methyl histidine	0.5	0.2	NS	NS	NS	3.2	1.1	NS	NS	NS
4-aminobutyric acid	555.2	1291.4**	NS	973.5**	NS	622.1	847.1	NS	NS	NS
4-hydroxyproline	78.7	72.9	NS	NS	NS	87.1	35	NS	NS	NS
α -aminoadipic acid	7.4	3.8	NS	NS	NS	1.6	2.2	NS	NS	NS
α -aminobutyric acid	123.4	115.2	NS	NS	NS	88.2	128.4	NS	NS	NS
agmatine	69.9	48.3	NS	NS	NS	56.6	49.4	NS	NS	NS
agmatine	65.3	48.5	NS	NS	NS	76	51.1	NS	NS	NS
aminopimelic acid	1	0.5	NS	NS	NS	0.3	0.4	NS	NS	NS
arginine	54	46.2	NS	NS	NS	62.2	48.8	NS	NS	NS
aspartic acid	721.7	703.2	NS	NS	NS	468.9	373	NS	NS	589.8*
β -aminoisobutyric acid	1.9	2.1	NS	NS	NS	1.5	2.2	NS	NS	NS
cadaverine	277	135.7*	NS	NS	NS	0.7	0.4	NS	NS	NS
citrulline	830.1	610.8	NS	NS	NS	317.3	395.2	NS	NS	NS
cystathionine	18.6	17.8	7.6*	6.1**	NS	5.4	9.4	NS	NS	NS
cysteine	4.7	5.3*	4**	NS	NS	4.3	5.8	NS	NS	NS
glutamic acid	21195.4	15715	NS	NS	NS	11381.7	8000.2	NS	NS	NS
glutamine	137.7	111.1	NS	NS	NS	110.4	85.1	NS	NS	NS
glycine	8498.8	6788.8	NS	NS	NS	3195.3	3770.8	NS	NS	NS

glycine-proline	9.9	5.3	8.6*	NS	13.9**	9.2	6	NS	NS	NS
histidine	310.6	141.4	327.6*	349*	NS	120.2	220.8	NS	NS	NS
hydroxylysine	44.7	31.1	NS	NS	NS	55.7	38.7	NS	NS	NS
isoleucine	994.7	914.5	NS	NS	NS	199	218.3	NS	NS	NS
leucine	1713.8	1313.6	NS	NS	NS	624.5	945.1	NS	NS	NS
lysine	2207.3	2478.6	NS	NS	NS	255.2	524.3**	345.9**	NS	393.7*
methionine	52.5	54.9	NS	NS	NS	7.6	5.4	NS	18.2*	NS
ornithine	109	84.4	103.2*	127.8*	141.9**	58	90.5**	NS	NS	124.1*
phenylalanine	1005.7	599.7*	NS	NS	1065.1**	238.4	351.2*	NS	261.2**	269.6**
proline	608.8	608.1	NS	NS	NS	331.2	273.6	NS	NS	NS
putrescine	1876.4	1877.4	NS	NS	NS	446.4	2217.9**	731.3**	1041.7**	919.6**
sarcosine	8.4	12	NS	NS	NS	124.1	85.1	NS	NS	NS
serine	3092	3530.8	2063.5*	NS	NS	1524	1894.8	NS	NS	NS
spermidine	286	254	NS	NS	NS	52.5	225.5**	93.8**	141.6*	90.3**
spermine	0	0	NS	NS	NS	0	0.1	NS	NS	NS
thiaproline	91.5	231.4	NS	NS	NS	152.5	156.5	NS	NS	NS
threonine	499.2	663.9	NS	508.2*	NS	273	285.6	NS	NS	NS
tryptophan	11.4	7.1	NS	NS	10**	6.9	6.7	NS	NS	11.8*
tyrosine	264.2	124.1*	148.9*	NS	288.6**	134.2	49.2*	107.6**	68.8*	167.1**
valine	3760.4	2798	NS	NS	NS	1331.7	1879.9	NS	NS	NS