

Supplementary Materials

Table S1. Primers of genes used for qRT-PCR.

Primer name	Sequence (5'–3')
<i>ryhB</i> -1-F	AAAAAGAAGTAGACAAGTGCGAATG
<i>ryhB</i> -1-R	AGCAATGTGAGCAATGTCGTG
<i>ryhB</i> -2-F	GCGGCTGAAAAAGACCATGA
<i>ryhB</i> -2-R	CCTGTTCGGTACGACATTGCT
<i>ssaI</i> -F	AACCGAGCCAGGAGCAAATTA
<i>ssaI</i> -R	TTATTAAGCGCTTGCGATGCT
<i>sseA</i> -F	GGCAGAGAGCCCCAAAAAGA
<i>sseA</i> -R	CCGGGGCTTGAGCATTAAGT
<i>pagC</i> -F	TATCCGAGGGGTAAATGTGA
<i>pagC</i> -R	CAGCGCGTATAACGAAAAAT
<i>sodC</i> -F	GTAGAGATGAATCTGGTGACG
<i>sodC</i> -R	TTCATGCTTGCCGGTATTT
<i>mgtC</i> -F	TGTCTCTGGTATTGGCTTTC
<i>mgtC</i> -R	CGGGAGTTGATTTATACGCT
<i>yaeB</i> -F	GAAATTAGGCAGTCTGGATCT
<i>yaeB</i> -R	CGTTAAAATCATGCAGCCAG
<i>pocR</i> -F	GCCTCACATGTCTATCTCAG
<i>pocR</i> -R	AGACTTTGCAAAAATAGCTGG
<i>hns</i> -F	TTCTGAACAACATCCGTACT
<i>hns</i> -R	CTATTCAGCAGTTCATTCGG
<i>arcC</i> -Fwd	ATTGCTGGCGGAGCAGATAG
<i>arcC</i> -Rev	CCGCTTCACATACCCGCTTA
<i>nark</i> -Fwd	GGTATCCTCATCGTGCCCTG
<i>nark</i> -Rev	TATTGCCCAGACCACCGTTC
<i>nirB</i> -Fwd	ATCATCGCCACGGAGAAAGG
<i>nirB</i> -Rev	GGGTCAGTTTGTGCGGCAGTA
<i>nirD</i> -Fwd	CGCGATTTTCCGTCCTTACC
<i>nirD</i> -Rev	CGCAATCAGTCCACGAGACA
<i>narG</i> -Fwd	ACCAACGTTACGAGCTGAT
<i>narG</i> -Rev	AGGAAGTTGAGCGCCTTCTC
<i>sipA</i> -Fwd	AGGCGAAGACGCTACTGATG
<i>sipA</i> -Rev	GCTTCGCTTCCGCTTTCTTT
<i>sopE</i> -Fwd	AGGTAGCGCGAGTAAAGACC
<i>sopE</i> -Rev	CGTCATTCTTGTTGCTCCG
<i>tolQ</i> -Fwd	CGGGTATTGCCGAAGCATTG
<i>tolQ</i> -Rev	TACCCCTTGTTGCTTTTCGCT
<i>hscA</i> -Fwd	CGCTATCCGCATCTGCCTTA
<i>hscA</i> -Rev	CGAAATAGGCGGGAACGGTA
<i>nrdB</i> -Fwd	TCTGCTGGCGTTACTGGTTT
<i>nrdB</i> -Rev	CATCACCACCATGACCACCA

Table S2. Primers for construction of recombinant plasmids.

Primer name	Sequence (5'-3')
<i>ssaI</i> -pXG-10SF-Fwd	GAGATACTGAGCACATGCATAATATTCTGATATGAGCGTAGTGC
<i>ssaI</i> -pXG-10SF-Rev	AGCCAGCGGATCCGCTAGCTGCCATGTTATCCAGCTT
<i>sseA</i> -pXG-10SF-Fwd	GAGATACTGAGCACATGCATTTTCATCCCTGGCCTTAACG
<i>sseA</i> -pXG-10SF-Rev	AGCCAGCGGATCCGCTAGCCCTTTTGTTCCTGACGG
pXG-10SF-seq-Fwd	CGCCAGATATCGACGTCT
pXG-10SF-seq-Rev	AAAGACAGGCACCTCTCC

Table S3. Summary about total reads successfully aligned to reference genome.

Sample name	Total reads	Successfully aligned reads	Ratio
WT1	4474322	4305857	96%
WT2	5471969	5207970	95%
WT3	5579891	5293677	95%
Δ <i>ryhB</i> -1-1	5476167	5256237	96%
Δ <i>ryhB</i> -1-2	5814310	5544776	95%
Δ <i>ryhB</i> -1-3	5756354	5526504	96%
Δ <i>ryhB</i> -2-1	5969797	5712121	96%
Δ <i>ryhB</i> -2-2	6090677	5817934	96%
Δ <i>ryhB</i> -2-3	5725666	57554	96%
Δ <i>ryhB</i> -1 Δ <i>ryhB</i> -2-1	5767623	5505745	95%
Δ <i>ryhB</i> -1 Δ <i>ryhB</i> -2-2	6323152	6000992	95%
Δ <i>ryhB</i> -1 Δ <i>ryhB</i> -2-3	6007256	5713601	95%

Table S4. Information of 13 candidate target genes screened by RNA-Seq.

Gene name	Product	$\log_2(\text{FC})$		
		WT vs. Δ <i>ryhB</i> -1	WT vs. Δ <i>ryhB</i> -2	WT vs. Δ <i>ryhB</i> -1/ Δ <i>ryhB</i> -2
<i>sipA</i>	pathogenicity island 1 effector protein	-1.043961057	-0.449383288	-0.492440637
<i>sopE</i>	guanine nucleotide exchange factor SopE	-1.30455987	NS	-1.284453795
<i>hns</i>	global DNA-binding transcriptional dual regulator H-NS	1.314465854	1.825069442	1.278914736
<i>ssaI</i>	pathogenicity island protein	2.40329259	1.563195229	1.749635333
<i>sseA</i>	pathogenicity island protein	1.749390717	1.269857594	1.708645919
<i>arcC</i>	carbamate kinase	NS	NS	-1.280819577
<i>narK</i>	NarK family nitrate/nitrite MFS transporter	-1.372546441	-1.255806586	-1.62991254
<i>nirB</i>	nitrite reductase large subunit	-1.269562961	-1.118081493	-1.325694806
<i>nirD</i>	nitrite reductase small subunit NirD	NS	NS	-1.268219173
<i>narG</i>	carbamate kinase	NS	NS	-1.204395098
<i>tolQ</i>	colicin uptake protein TolQ	0.355563326	0.33111866	0.541312818
<i>hscA</i>	chaperone protein HscA	0.498653077	0.712040375	0.614042734
<i>mrdB</i>	cell wall shape-determining protein	0.664299108	0.765219803	0.466340675

WT: wild-type; NS: not significant.

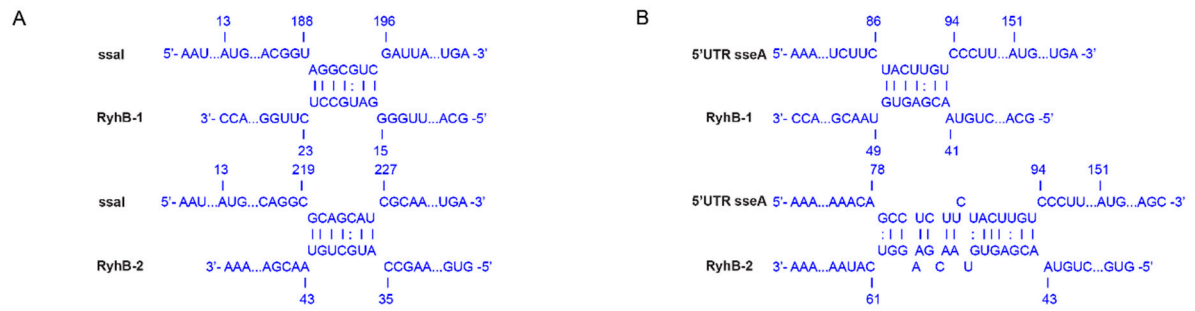


Figure S1. Interaction site prediction between RyhB-1/RyhB-2 and putative target genes. **(A)** Prediction between RyhB-1/RyhB-2 and *ssal*. CDS region of *ssal* is predicted to form base pairs with RyhB-1 and RyhB-2. **(B)** Prediction between RyhB-1/RyhB-2 and *sseA*. 5' UTR of *ssal* is predicted to form base pairs with RyhB-1 and RyhB-2.