Supporting Information

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SI Methods

Plasmid Construction. Plasmids are listed in Table S3, and primers are listed in Table S4. DNA extractions, purifications, electrophoresis, PCR, and sequencing were carried out as described (1–3). All DNA cloning was carried out by using *E. coli* EPI300, which was supplemented with 10% (wt/vol) arabinose when propagating $oriV^{RK2}$ plasmids.

pSDZ. The GFP-fusion cassette from pJRGFUS was amplified by using primers 1 and 2, and the product was digested with BamHI and treated with T4 polynucleotide kinase. This BamHI–Blunt fragment was ligated with pFAJ1700 DNA that had been sequentially digested with HindIII, treated with T4 DNA polymerase, and digested with BamHI. The resulting plasmid was digested with Asp-718 and BamHI and the *lacI–lacO* region, amplified from pSRKKm by using primers 3 and 4 and digested with Asp-718 and BamHI, was cloned into it. This plasmid was digested with Asp-718 and the *lacZ* gene, amplified from pFUS2 by using primers 5 and 6 and digested with Asp-718, was cloned into it. This plasmid was digested with XhoI, and the *rrnB1* terminator, amplified from *E. coli* BL21(DE3)pLysS DNA by using primers 7 and 8, was cloned as an XhoI–SalI fragment (Fig. S2).

pSDrdfS–lacZ. The *rdfS* gene was amplified from *M. loti* genomic DNA by using primers 9 and 10 and cloned into pSDZ as a HindIII–PstI fragment.

p172171rdfS–lacZ. pSDrdfS–lacZ was digested with EcoRI and HindIII, and the *msi172–msi171* region, amplified from *M. loti* genomic DNA with primers 11 and 12, was cloned as an MfeI–HindIII fragment.

pFseArdfS–lacZ and T231C and C232A–C234G derivatives of p172171rdfS–lacZ. Cloning was carried out as for p172171rdfS–lacZ, except that the PCR product cloned was created by using overlap-extension PCR, as described (1), using complementary primers 13 and 14, 15 and 16, and 17 and 18, respectively.

pMAL172171. The *msi172–msi171* PRF region was amplified by using primers 27 and 28 and cloned as an MfeI–HindIII fragment into pMAL-c2 cut with EcoRI and HindIII.

pTRGmsi172. *msi172* was amplified by using primers 31 and 32 and cloned into pTRG as a NotI–XhoI fragment.

pTRGmsi171. *msi171* was amplified by using primers 29 and 30 and cloned into pTRG as a NotI–EcoRI fragment.

pTRGmsi172–msi171 and pTRGfseA. Primers 33 and 34 were used to amplify *msi172–msi171* from p172171rdfS–lacZ and pFseArdfS–lacZ, respectively, and were cloned as MunI–SpeI fragments in pTRG.

pUC172171, pUC172171+ve, and pUC172171-ve. Complementary oligonucleotide pairs 35 and 36, 37 and 38, and 39 and 40 were annealed by cooling from 99 °C to room temperature, after which they were treated with T4 polynucleotide kinase. The HindIII- and EcoRI-compatible overhangs on the complementary oligonucleotides facilitated directional cloning into pUC19 digested with HindIII and EcoRI for each of the three plasmids.

MBP Purification. A 10-mL LB broth culture of *E. coli* EPI300 carrying plasmid pMAL172171 was used to seed a 1-L volume of 2xTY medium containing ampicillin. This culture was grown at 37 °C with shaking, and after it reached an OD_{600} of 0.4–0.7, 1 mM IPTG was added to induce expression from pMAL172171. After 2 h, cells were recovered by centrifugation for 20 min at 10,000 × *g* and stored at −80 °C. The pellet, together with 10 g of aluminum(III) oxide was ground with mortar and pestle and then suspended in extraction buffer [20 mM Tris (pH 8), 250 mM NaCl, 1 mM EDTA, 1 mM DTT, 1 mM PMSF, and 1 μg/mL DNase I]. The lysate was cleared by centrifugation for 20 min at 10,000 × *g*. Cleared lysate was mixed with 4 mL of amylose resin (prewashed with extraction buffer) and incubated at 4 °C on a rotating wheel. The resin was placed in a column and washed with 3 × 10 mL of column buffer [20 mM Tris (pH 8), 250 mM NaCl, 1 mM EDTA, and 1 mM DTT] before being eluted with 12 mL of column buffer containing 10 mM maltose. One-milliliter fractions were collected and analyzed by reducing SDS/PAGE. Densitometry analysis was carried out by using an Odyssey Fc dual-mode imaging system (LI-COR Biosciences) and ImageJ software (4).

LTQ Orbitrap MS.

Concentrated peptides were loaded onto a custom-made nanoflow emitter tip column (75-μm ID silica tubing packed with 3-μm C18 beads at a length of 12 cm) and separated by a gradient of 5% (wt/vol) solvent B (0.2% formic acid in acetonitrile) in solvent A (0.3% formic acid in water) to 30% (wt/vol) solvent B in solvent A over 15 min, followed by an increase to 95% (wt/vol) solvent B in solvent A over 7 min. Spectra were acquired in data-dependent mode using 1 Orbitrap survey scan at a resolution of 60,000, followed by five collision-induced dissociation tandem mass spectrometry scans acquired in the LTQ ion trap analyzer. Raw data were processed through the Proteome Discoverer software (Version 1.4; Thermo Scientific) using default settings for the generation of peak lists. A custom sequence database containing all possible peptides that could be produced following a PRF at any position downstream of the A437 trypsin site was created. Spectra were then searched against this database by using the SEQUEST HT (Thermo Scientific) and Mascot (Mascot in-house server; Version 2.5; Matrix Science) search engines. In addition, spectra were searched against the SwissProt amino acid sequence database (458,075 sequence entries) to confirm that significant peptide spectrum matches of frameshift products were not matching any other sequences by chance.

1. Ramsay JP, Sullivan JT, Stuart GS, Lamont IL, Ronson CW (2006) Excision and transfer of the *Mesorhizobium loti* R7A symbiosis island requires an integrase IntS, a novel recombination directionality factor RdfS, and a putative relaxase RlxS. *Mol Microbiol* 62(3):723–734.
2. Ramsay JP, et al. (2009) A LuxRI-family regulatory system controls excision and transfer of the *Mesorhizobium loti* strain R7A symbiosis island by activating expression of two conserved hypothetical genes. *Mol Microbiol* 73(6):1141–1155.
3. Ramsay JP, et al. (2013) A widely conserved molecular switch controls quorum sensing and symbiosis island transfer in *Mesorhizobium loti* through expression of a novel antiactivator. *Mol Microbiol* 87(1):1–13.
4. Schneider CA, Rasband WS, Eliceiri KW (2012) NIH Image to ImageJ: 25 years of image analysis. *Nat Methods* 9(7):671–675.

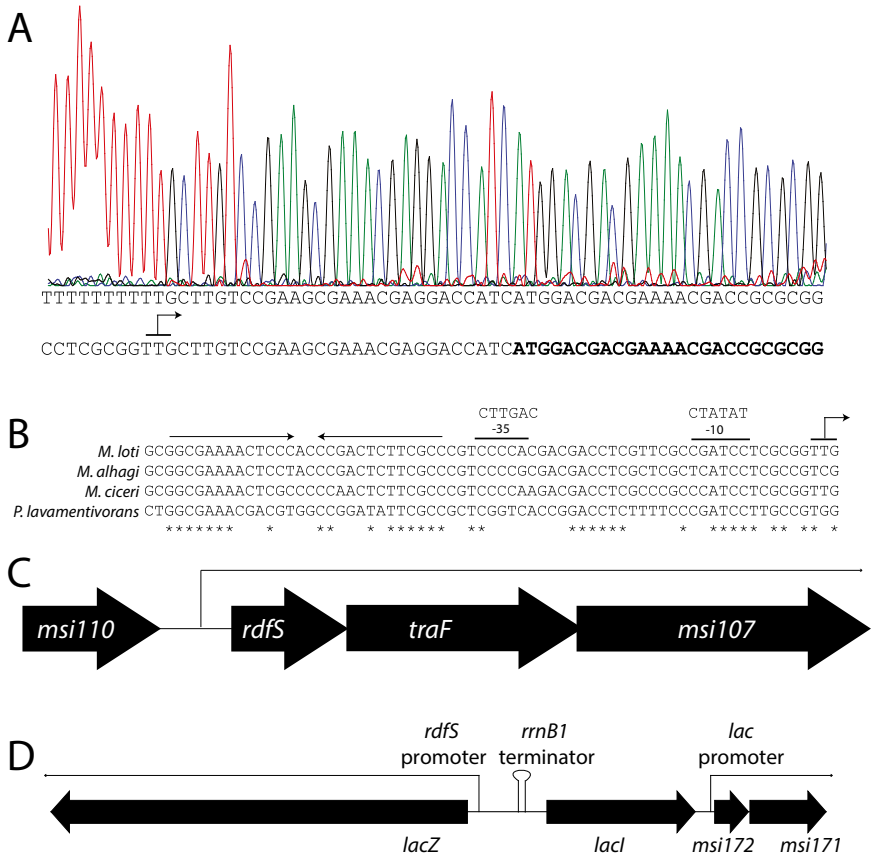

Fig. S1. Mapping the transcriptional start site of *rdfS*. (*A*) Reverse-complemented sequence chromatogram of PCR-amplified cDNA. The poly-T sequence indicates that dATP attached to the 3′ of cDNA during 5′ RACE reactions. The corresponding DNA sequence is aligned below. The transcriptional start site was localized to the one of three nucleotides (TTG) shown by a black bar and arrow. The *rdfS* ORF is shown in bold. (*B*) Alignment of the promoter regions upstream of *rdfS* homologs in *M. alhagi*, *M. ciceri*, and *P. lavamentivorans*. Completely conserved nucleotides are indicated with asterisks, and inverted arrows highlight a conserved inverted-repeat sequence. The rhizobial consensus −35 and −10 sequences (1) are shown above their approximate corresponding positions in the *rdfS* promoter (*C*) Genetic organization of the *rdfS* operon and position of the *rdfS* promoter (arrow). (*D*) Map of cloned genes in p172171rdfS–lacZ.

1. MacLellan SR, MacLean AM, Finan TM (2006) Promoter prediction in the rhizobia. *Microbiology* 152(Pt 6):1751–1763.

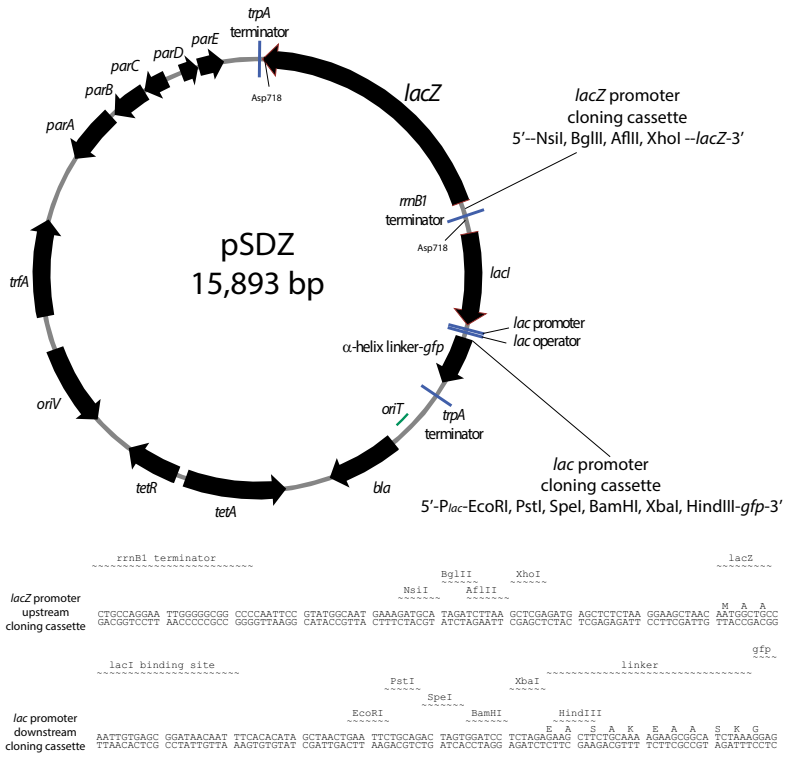

Fig. S2. Map of multipurpose cloning vector pSDZ. Plasmid pSDZ was constructed by using the stable low-copy number RK2 replicon plasmid pFAJ1700 (1) as a backbone, which carries Ap^R and Tet^R and an *oriT* to facilitate conjugative mobilization. pSDZ carries a linker-*gfp* gene that lacks a start codon or RBS, for construction of protein fusions with GFP. Upstream of the *gfp* gene is a cloning cassette shown below the plasmid map. This cloning cassette is positioned downstream of the LacI-binding site and *lac* promoter, making the promoter lactose or IPTG-inducible. The *lacI* gene is also carried upstream of this site. A second cloning cassette is positioned upstream of a promoterless *lacZ* gene and downstream of the *rrnB1* transcriptional terminator. The cassette upstream of *lacZ* facilitates cloning of promoter sequences for analysis of expression using β-galactosidase assays.

1. Dombrecht B, Vanderleyden J, Michiels J (2001) Stable RK2-derived cloning vectors for the analysis of gene expression and gene function in gram-negative bacteria. *Mol Plant Microbe Interact* 14(3):426–430.

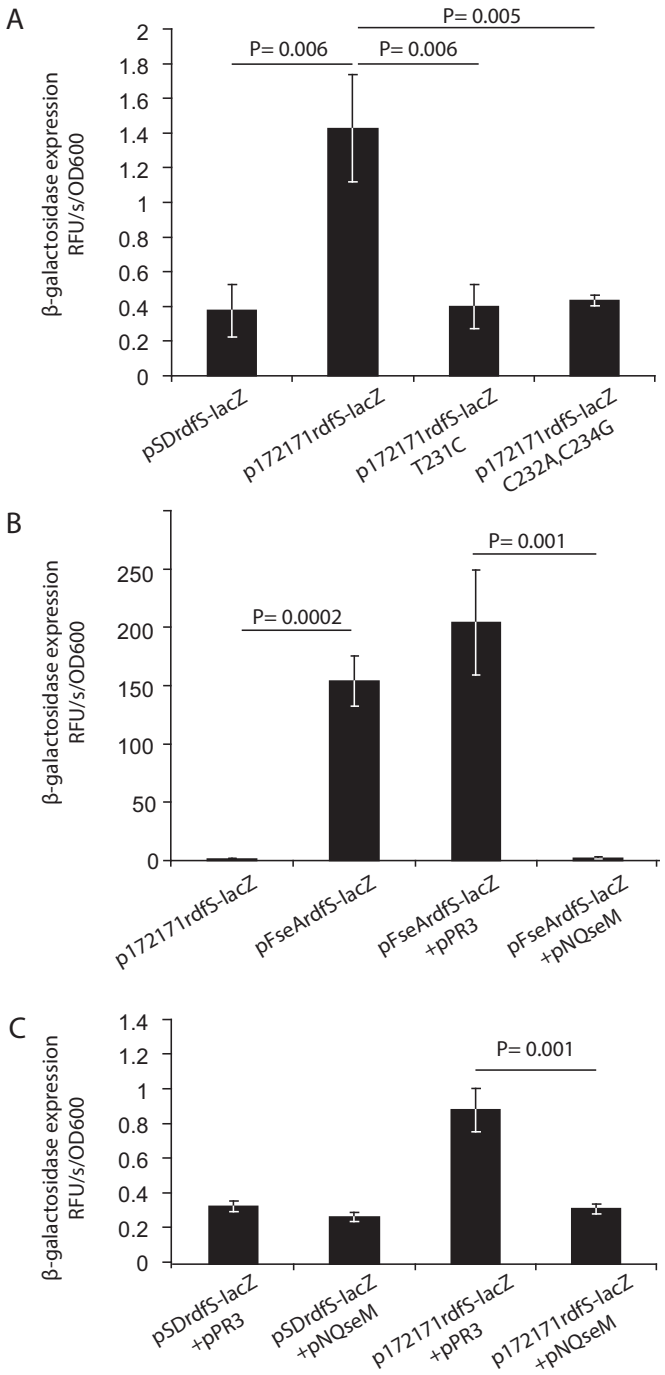

Fig. S3. Activation of the *rdfS* promoter by *msi172–msi171*. (*A*) β-galactosidase assays of *M. loti* R7ANS cells carrying p172171rdfS–lacZ and derivatives carrying mutations in *msi172–msi171* that abolish *rdfS* expression. (*B*) β-galactosidase expression from pFseArdfS-pro, which contains *msi172–msi171* carrying a frameshift deletion mutation that fuses the *msi172* and *msi171* ORFs, and repression of expression from pFseArdfS–lacZ by the constitutive *qseM* expressing plasmid pNQseM. (*C*) Repression of expression from p172171rdfS–lacZ by plasmid pNQseM.

A

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Frame 0  Q  W  G  L  L  F  R  K  L  V  E  A  STOP
Frame +1                F  A  S  S  L  R  P  D  A -LacZ-alpha
5'-AGCTTGCAGTGGGGGCTCCTGTTTCGCAAGCTCGTTGAGGCCTGATGCG-3'

Frame 0  Q  W  G  L  L  F  A  S  S  L  R  P  D  A -LacZ-alpha
5'-AGCTTGCAGTGGGGGCTCCTGTTCGCAAGCTCGTTGAGGCCTGATGCG-3'

Frame 0  Q  W  G  L  L  F  R  K  L  V  E  A  STOP
Frame +1                F  A  S  S  STOP         -LacZ-alpha
5'-AGCTTGCAGTGGGGGCTCCTGTTTCGCAAGCTCGTAGAGGCCTGATGCG-3'
```

B

	β-galactosidase activity (RFU/s/Ab_{600})	Standard deviation
pUC19 (empty vector)	685.81	43.53
pUC172171	31.24	9.96
pUC172171+ve	219.37	3.77
pUC172171-ve	3.44	0.82

Fig. S4. PRF-dependent expression of LacZ-α on pUC19. (*A*) DNA sequence of oligonucleotides cloned into pUC19 to enable measurement of ribosomal frameshifting from the *msi172* PRF site. The PRF region is highlighted in bold, and the mutated codons in the positive and negative control constructs are underlined. (*B*) β-galactosidase assays of *E. coli* carrying the pUC19 constructs outlined in *A*.

Table S1. Sequences of PRF regions present on ICE*Ml*Sym^{R7A}-like elements

SRV.TGG.GGN.NTN.NNN.TTT.CSY PRF sites on ICE*ML*Sym^{R7A}-like elements	
*Mesorhizobium loti*_R7A_ICE*Ml*Sym^{R7A}*	TCGCTCGATCTCACG**CAGTGGGGGCTCCTGTTTCGC**AAGCTCGTTGAGGCCT
Xanthobacter autotrophicus Py2_ICE1	AGGAGCACCGCCCCG**CAATGGGGATTGCGCTTTCCT**GATCGATCCCGATCGA
*Bradyrhizobium_BTAi1*_ICE29	GTGCTGGTGCGGCCG**CGCTGGGGGTTGCGCTTTCGC**GATCGACCCGTCGCTC
*Chelativorans_sp.*_BNC1_ICE1	GAGGTGATCCAGCGC**CGCTGGGGGTTGCGATTTCCC**GGCGAGGCCGTCCCAT
*Chelativorans_sp.*_BNC1_ICE2	GAGGCGACGAGGTCT**CGATGGGGGCTGCGATTTCCT**GTTGTCGCCGGCCCTC
*Chelativorans*_sp._BNC1_ICE3	GAGGAGTTCTCGAAG**CGCTGGGGCTTGCGATTTCCC**GTGTGATCCCGCAACC
Rhodopseudomonas. palustris NC_008435	ATCCCCTTCAGGAGC**CAATGGGGCCTCTCCTTTCGC**CGCTGATCCGACGTTG
Rhodopseudomonas. palustris _Bis18_ICE1	ATACCCTTCAGGAGC**CAATGGGGCCTCTCCTTTCGC**CGCTGATCCGACGTTG
Rhodopseudomonas. palustris _Bis18_ICE2	GAAGCCTTTTCCGAG**CGCTGGGGGTTAAGGTTTCCC**GCGAGACCCGCGCCAG
*Paracoccus.denitrificans*_PD1222_ICE1	GCCCGGATGGCCCGC**CGCTGGGGCCTGATGTTTCCC**GGTCGATCCCGCACTT
Paracoccus.denitrificans _PD1222_ICE2	GATCAGGTCGGCCGT**CGATGGGGTCTATGCTTTCCC**GATACGGCCTGGTCTG
Paracoccus.denitrificans _PD1222_ICE3	CTTCGGTTCCGGAAT**CGCTGGGGGCTTACGTTTCCC**GGTCCGGCCCGGACTG
*M. loti*_MAFF303099	TCGCTCGATCTCACG**CAGTGGGGGCTCCTTTTTCGC**AGGCTCGTTGAGGCGC
Oceanicaulis_alexandrii	GGCGGCGCTCCAGCA**GACTGGGGGTTACGATTTCGC**TGTAAACCCACAGCTC
Single ORF (no PRF) sequences on ICE*Ml*Sym^{R7A}-like elements	
*Sphingomonas_sp.*_SKA58_ICE1	GACTCGAGCATAGTG**CGCTGGGGCTGCCTAAACCTG**CCGGACGCCGCTCTTC
*Parvibaculum lavamentivorans*_DS-1_ICE1	ATGTCCGAGGCACGG**GAATGGGGCTTGCTCCTTTTC**GAAACGCCCGATCGCG
Sequence where *msi172* and *msi171* are +1 nucleotide out-of-frame but the ICE*ML*Sym^{R7A} PRF motif is absent	
Bradyrhizobium_USDA_110	ACCAGTTTCGCACGT**CGATGGAGGTTGCGGTGTCGC**CCATGACCCCGCCTCT
Mesorhizobium sp.† *msi172* homologs carrying a TTT.T type PRF	
LSHC440B00,LSHC440A00,LSJC285A00	ACTGTTGATCTCTCG**CGCTGGGGCCTTCTGTTTTGC**AAGCTCAAAAGGAACT
LNJC391B00, LSJC277A00	ACTGTTGATCTCTCG**CGGTGGGGCCTTCTGTTTTGC**AAGCTCAAAAGGAACT
L2C054A000	ACTGTCGATCTTTCG**CGGTGGGGCCTTCTGTTTTGC**AAGCTCAAAAGGAACT
Mesorhizobium plurifarium (emb|CDX12642.1)	GAAACCGATCTTTCC**CTCTGGGGGGTTCTCTTTTGC**AAGGTCGCTCGAGAGT
L48C026A00	ACTGTTGATCTCTCG**CGGTGGGGCCTTCTGTTTTGC**AAGCTCGAAAGGAACT
LSHC414A00	ACTGTCGATCTTTCG**CCGTGGGGCCTTCTGTTTTGC**AAGCTCTATCGAAACT

*Accession numbers and annotations of ICE*Ml*Sym^{R7A}-like ICEs have been published (1).
†Accession numbers available from Bioproject PRJNA171524 unless otherwise stated.

1. Ramsay JP, et al. (2013) A widely conserved molecular switch controls quorum sensing and symbiosis island transfer in *Mesorhizobium loti* through expression of a novel antiactivator. *Mol Microbiol* 87(1):1–13.

Table S2. Bacteriomatch II two-hybrid assay, cfu/mL

pTRG clone	GAL2P	vector	vector	*msi172–msi171*	*msi172–msi171*	*fseA*	*fseA*	*msi172*	*msi172*	*msi171*	*msi171*
pBT clone	LGF2	vector	*qseM*	vector	*qseM*	vector	*qseM*	vector	*qseM*	vector	*qseM*
Nonselective, cfu	1.57×10^{6}*	1.05×10^{6}	8.3×10^{5}	6.12×10^{6}	1.096×10^{7}	2.096×10^{7}	2.11×10^{7}	3.04×10^{7}	1.24×10^{7}	9.42×10^{6}	1.78×10^{7}
Selective, cfu	$\mathbf{5.28 \times 10^{3}}$	6×10^{1}	4×10^{1}	3.2×10^{2}	$\mathbf{2.65 \times 10^{6}}$	4.8×10^{2}	$\mathbf{4.44 \times 10^{6}}$	9.6×10^{2}	$\mathbf{5.44 \times 10^{6}}$	4×10^{1}	1.39×10^{3}
Ratio	$\mathbf{3.35 \times 10^{-3}}$	5.71×10^{-5}	4.82×10^{-5}	5.23×10^{-5}	$\mathbf{2.42 \times 10^{-1}}$	2.29×10^{-5}	$\mathbf{2.1 \times 10^{-1}}$	3.1×10^{-5}	$\mathbf{4.37 \times 10^{-1}}$	4.25×10^{-6}	7.79×10^{-5}

*Values are from a single representative experiment. All combinations were repeated at least twice, and replicates produced similar results. Values indicating positive interactions are in bold.

Table S3. Strains and plasmids

Strain/plasmid	Description	Source
M. loti		
R7A	Field reisolate of ICMP 3153; wild-type symbiotic strain	Ref. 1
R7ANS	Nonsymbiotic derivative of R7A; lacks ICE*Ml*Sym^{R7A}	Ref. 2
E. coli		
BL21(DE3)pLysS	F$^-$ *ompT gal dcm lon hsdS$_B$*(r$_B^-$ m$_B^-$) λ(DE3) pLysS, Cm^R	Ref. 3
DH5α	*fhuA2 lac*(del)U169 *phoA* glnV44 Φ80′ *lacZ*(del)M15 *gyrA96 recA1 relA1 endA1 thi-1 hsdR17*	Ref. 4
EPI300	F$^-$ *mcrA* Δ(*mcrCB-hsdSMR-mrr*) (Str^R) Φ80d*lacZ*ΔM15 Δ*lacX74 rec*A1 *end*A1 *araD*139 Δ(*ara, leu*)7697*galU galK* λ$^-$ *rpsL nupG trfA ton*A *dhfr*	Epicenter
ST18	S17 λ*pir* Δ*hemA*	Ref. 5
BacterioMatch II	Δ(*mcrA*)*183* Δ(*mcrCB-hsdSMR-mrr*)*173 endA1 hisB supE44 thi-1 recA1 gyrA96 relA1 lac* [F′ *laqI^q HIS3 aadA* Kan^R]	Agilent
Plasmids		
pFAJ1700	Broad-host-range plasmid, *oriV^{RK2}* Tc^R, Ap^R	Ref. 6
pPR3	pVS1/p15a replicons, carries *nptII* promoter, Km^R	Ref. 7
pUC19	High-copy-number cloning vector, Ap^R	NEB
pNqseM	pPR3 carrying the *qseM* gene cloned downstream of the *nptII* promoter, amplified using primers msi1703clone and msi170ATG and cloned as a BamHI fragment	Ref. 8
pBT	Bacteriomatch II bait cloning vector carrying λcI, Cm^R	Agilent Technologies
pTRG	Bacteriomatch II target cloning vector carrying RNA polymerase α fragment, Tc^R	Agilent Technologies
pTRG-Gal11^P	Bacteriomatch II positive control plasmid, Tc^R	Agilent Technologies
pBT-LGF2	Bacteriomatch II positive control plasmid, Cm^R	Agilent Technologies
pBTqseM	pBT carrying a cI-QseM fusion, *qseM* was amplified using primers 1702HNotI5′ and 1702HNotI3′ and cloned as a NotI fragment	Ref. 8
pSRKKm	*lacI-lacO* expression vector, Km^R	Ref. 9
pJRGFUS	GFP fusion cassette vector Ap^R	Ref. 10
pFUS2	*lacZ* fusion vector Gm^R	Ref. 11
pMAL-c2	MBP expression vector, Ap^R	NEB
pSDZ	Derivative of pFAJ1700 carrying *lacZ* gene from pFUS2 and *lacI-lacO* from pSRKKm and the GFP fusion cassette from pJRGFUS.	This study
pSDrdfS-lacZ	pSDZ carrying the *rdfS* promoter region	This study
p172171rdfS-lacZ	pSDrdfS-lacZ carrying *msi172-msi171*	This study
pFseArdfS-lacZ	p172171rdfS-lacZ with a T229 nucleotide deletion in *msi172*	This study
p172171rdfS-lacZ^{T231C}	p172171rdfS-lacZ with a T231C nucleotide substitution in *msi172*	This study
p172171rdfS-lacZ$^{C232A.C234G}$	p172171rdfS-lacZ with C232A and C234G nucleotide substitutions in *msi172*	This study
pMAL172171	pMAL-c2 carrying the PRF site of *msi172-msi171*	This study
pTRGmsi172	pTRG carrying *msi172*	This study
pTRGmsi171	pTRG carrying *msi171*	This study
pTRGmsi172-msi171	pTRG carrying *msi172-msi171*	This study
pTRGfseA	pTRG carrying *msi172-msi171* from pFseArdfS-lacZ	This study
pUC172171	pUC19 carrying cloned oligonucleotides 35 and 36	This study
pUC172171+ve	pUC19 carrying cloned oligonucleotides 37 and 38	This study
pUC172171-ve	pUC19 carrying cloned oligonucleotides 39 and 40	This study

1. Sullivan JT, Patrick HN, Lowther WL, Scott DB, Ronson CW (1995) Nodulating strains of *Rhizobium loti* arise through chromosomal symbiotic gene transfer in the environment. *Proc Natl Acad Sci USA* 92(19):8985–8989.
2. Ramsay JP, Sullivan JT, Stuart GS, Lamont IL, Ronson CW (2006) Excision and transfer of the *Mesorhizobium loti* R7A symbiosis island requires an integrase IntS, a novel recombination directionality factor RdfS, and a putative relaxase RlxS. *Mol Microbiol* 62(3):723–734.
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5. Thoma S, Schobert M (2009) An improved *Escherichia coli* donor strain for diparental mating. *FEMS Microbiol Lett* 294(2):127–132.
6. Dombrecht B, Vanderleyden J, Michiels J (2001) Stable RK2-derived cloning vectors for the analysis of gene expression and gene function in gram-negative bacteria. *Mol Plant Microbe Interact* 14(3):426–430.
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8. Ramsay JP, et al. (2013) A widely conserved molecular switch controls quorum sensing and symbiosis island transfer in *Mesorhizobium loti* through expression of a novel antiactivator. *Mol Microbiol* 87(1):1–13.
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10. Tan H, et al. (2014) Comprehensive overexpression analysis of cyclic-di-GMP signalling proteins in the phytopathogen *Pectobacterium atrosepticum* reveals diverse effects on motility and virulence phenotypes. *Microbiology* 160(Pt 7):1427–1439.
11. Antoine R, et al. (2000) New virulence-activated and virulence-repressed genes identified by systematic gene inactivation and generation of transcriptional fusions in *Bordetella pertussis*. *J Bacteriol* 182(20):5902–5905.

Table S4. Primers

No.	Name	Sequence 5′–3′
1	GFPLinkerfwdBamHind	CGCCGGATCCTCTAGAGAAGCTTCTGCAAAAGAAGCGGCA
2	GFPlinkerrevBlunt	CTAATTAATCATTTGTATAGTTCAT
3	LacIfwd_KpnI	TATAGGTACCTGATTGACACCATCGAATGGTGCAAAACCT
4	lacIrevBamHI	TATGGATCCACTAGTCTGCAGAATTCAGTTAGCTATGTGTGAAATTGTTATCCGCT
5	LacZ5-KpnI_xhoI_SacI	TTAGGTACCCTCGAGATGAGCTCTCTAAGGAAGCTAACAATGGCT
6	lacZ3-KpnI	TATGGTACCTTATTTTTGACACCAGACCAACTGGTAATGGTAGCGA
7	5rrnB1_term_SalI	ATAGTCGACATGGCATGGATGAACTATACAAATAGGAATTA
8	3rrnB1_term_xhoI	TATCTCGAGCTTAAGATCTATGCATCTTTCATTGCCATACGGAA
9	rrnB1-rdfS-Fwd-hindIII	ATATAAGCTTCTCGACATGGCATGGATGA
10	rrnB1-rdfS-Rev-PstI	TATACTGCAGGATGGTCCTCGTTTCG
11	msi171_6H_HindIII_rev	TTAAGCTTAGTGGTGATGATGGTGATGGACGAGGAAATCCCGATA
12	pQe80_172-171_fwd_mfeI	GGATCAATTGAGAGGACAGGGATGGCTATGATAGGTAATGA
13	Msi172FS_3F	ACGCAGTGGGGGCTCCTGTTCGCAAGCTCGTTGAGGCCT
14	Msi172FS_5R	AGGCCTCAACGAGCTTGCGAACAGGAGCCCCCACTGCGT
15	FRAMESHIFTfwdprimer-T231C	TCTCACGCAGTGGGGGCTCCTGTTCCGCAAGCTCGTTGAGG
16	FRAMESHIFTrevprimer-T231C	CCTCAACGAGCTTGCGGAACAGGAGCCCCCACTGCGTGAGA
17	FRAMESHIFTfwdprimerC232A+C234G	TCTCACGCAGTGGGGGCTCCTGTTTAGGAAGCTCGTTGAGG
18	FRAMESHIFTrevprimerC232A+C234G	CCTCAACGAGCTTCCTAAACAGGAGCCCCCACTGCGTGAGA
19	rdfS_5_QPCR	GGCAGTCCGTTTCTGAACA
20	rdfS_3_QPCR	TCGATGTGGTAGCGGACATA
21	msi110_QPCR_5	GATCGGTTACGTCAACAAGCA
22	msi110_QPCR_3	CGGAAGTCGGGATGGTTGT
23	msi107_rev	CTCAACATCGCTAGCCATTGTGC
24	rdfS_SP1	GCTGCCGATCAGATGGATGGAA
25	rdfS_SP2	GGCCTTTCGACCAGTCGTCGAGT
26	rfdS_SP3	TCGTCGATGTGGTAGCGGACATA
27	pMAL_HindIII_msi171_3R	CGGCAAGCTTTGTCGACCGTCCGCCGTGATCAT
28	pMAL_MfeI_msi172_5F	GCGCCAATTGCGCTATACGCGGCACCTGACGCGA
29	Msi171 – Forward	ATATAAGCTTGCGGCCGCAGTGTTCTGGTGCTCCCGCCAAT
30	Msi171 – Reverse	TTTAAAGAATTCTTAGACGAGGAAATCCCGATAG
31	Msi172 – Forward	ATATAAGCTTGCGGCCGCAATGGCTATGATAGGTAATGACG
32	Msi172 – Reverse	AAATTTCTCGAGTCAGGCCTCAACGAGCTTGCGA
33	pTRG-171–171_Mun_I_fwd	ATATCAATTGGCGCTATGATAGGTAATGA
34	pTRG-172–171-rev	ATTAACTAGTTTAGTGGTGATGATGGTGA
35	172WT_Hin-Eco_fwd	AGCTTGCAGTGGGGGCTCCTGTTTCGCAAGCTCGTTGAGGCCTGATGCG
36	172WT_hin-Eco_Rev	AATTCGCATCAGGCCTCAACGAGCTTGCGAAACAGGAGCCCCCACTGCA
37	172POScon_hin_eco_fw	AGCTTGCAGTGGGGGCTCCTGTTCGCAAGCTCGTTGAGGCCTGATGCG
38	172POScon_hin_eco_re	AATTCGCATCAGGCCTCAACGAGCTTGCGAACAGGAGCCCCCACTGCA
39	172NEGcon_hin_eco_fw	AGCTTGCAGTGGGGGCTCCTGTTTCGCAAGCTCGTAGAGGCCTGATGCG
40	172NEGcon_hin_eco_re	AATTCGCATCAGGCCTCTACGAGCTTGCGAAACAGGAGCCCCCACTGCA