

TABLE S1. Bacterial strains and plasmids used in this work.

Strain or plasmid	Relevant genotype or phenotype	Source of reference
Strains		
<i>E. coli</i> strains		
DH5α	<i>supE44 ΔlacU169-(Ψ80lacZΔM15)hsdR1 recA1</i>	Bethesda Research Laboratories, Inc., Gaithersburg, U.S.A.
	<i>gyrA96 thi-1 relA1</i>	
S17-1	<i>Sm^r Sp^r; hdsR (RP4-2 kan::Tn7 tet::Mu, integrated in the chromosome)</i>	(1)
<i>B. japonicum</i> strains		
110spc4	<i>Sp^r; wild type</i>	(2)
6243	<i>Sp^r Km^r; Δbll3156-3157::aphII (same orientation)</i>	This work
Plasmids		
pGEM-T Easy	<i>Ap^r; cloning vector</i>	Promega Corporation, Madison, U.S.A.
pBSL15	<i>Ap^r Km^r; cloning vector</i>	(3)
pSUP202pol4	<i>Tc^r; (pSUP202) part of the polylinker from pBlueskript II KS+ between EcoRI and PstI</i>	(4)
pRJ6240	<i>Ap^r; (pGEM-T Easy) 623 bp amplicon of the bll3156 downstream region comprising the <i>B. japonicum</i> genome sequence from coordinate 3489814 to 3490437</i>	This work
pRJ6241	<i>Ap^r; (pGEM-T Easy) 636 bp amplicon of the bll3157 upstream region comprising the <i>B. japonicum</i> genome sequence from coordinate 3493519 to 3494155</i>	This work
pRJ6242	<i>Tc^r; (pSUP202pol4) 1259 bp EcoRI/NotI up- and downstream region of bll3156-bll3157</i>	This work
pRJ6243	<i>Tc^r Km^r; (pSUP202pol4) the <i>aphII</i> cassette flanked by the up-and downstream region of bll3156-bll3157</i>	This work

1. **Simon R, Priefer U, Pühler A.** 1983. Vector plasmids for *in vivo* and *in vitro* manipulation of gram-negative bacteria. Springer Verlag, Heidelberg, Germany.
2. **Regensburger B, Hennecke H.** 1983. RNA polymerase from *Rhizobium japonicum*. Arch Microbiol **135**:103-109.
3. **Alexeyev MF.** 1995. Three kanamycin resistance gene cassettes with different polylinkers. Biotechniques **18**:52-56.
4. **Fischer HM, Babst M, Kaspar T, Acuna G, Arigoni F, Hennecke H.** 1993. One member of a *groESL*-like chaperonin multigene family in *Bradyrhizobium japonicum* is co-regulated with symbiotic nitrogen fixation genes. EMBO J **12**:2901-2912.

TABLE S2. Specific nitrogenase activity (% C₂H₄ min⁻¹ g⁻¹) of *B. japonicum* wild type and 6243 mutant on soybean, mungbean, cowpea and siratro plants.

	Relevant genotype	Soybean	Mungbean	Cowpea	Siratro
110 <i>spc4</i>	Wild type	1.9 ± 0.7	1.9 ± 0.9	2.5 ± 0.7	1.7 ± 0.8
6243	$\Delta frc\text{-}oxc$	1.7 ± 1.2	2.2 ± 0.8	2.9 ± 1.7	1.2 ± 0.7