

NbPLC2s-VIGS

NbPLC2-1-VIGS
NbPLC2-2-VIGS

Figure S2 Nucleotide sequence of *NbPLC2-1* and *NbPLC2-2*

Alignment of nucleotide sequence of *NbPLC2-1* (Niben101Scf02221g00009) and *NbPLC2-2* (Niben101Scf00318g03011). Nucleotide sequence used for virus-induced gene silencing (NbPLC2s-VIGS, NbPLC2-1-VIGS, or NbPLC2-2-VIGS). Arrows indicated nucleotide sequence used for qRT-PCR (PLC2-1/2-2rtpF, PLC2-1rtpR and PLC2-2rtpR).

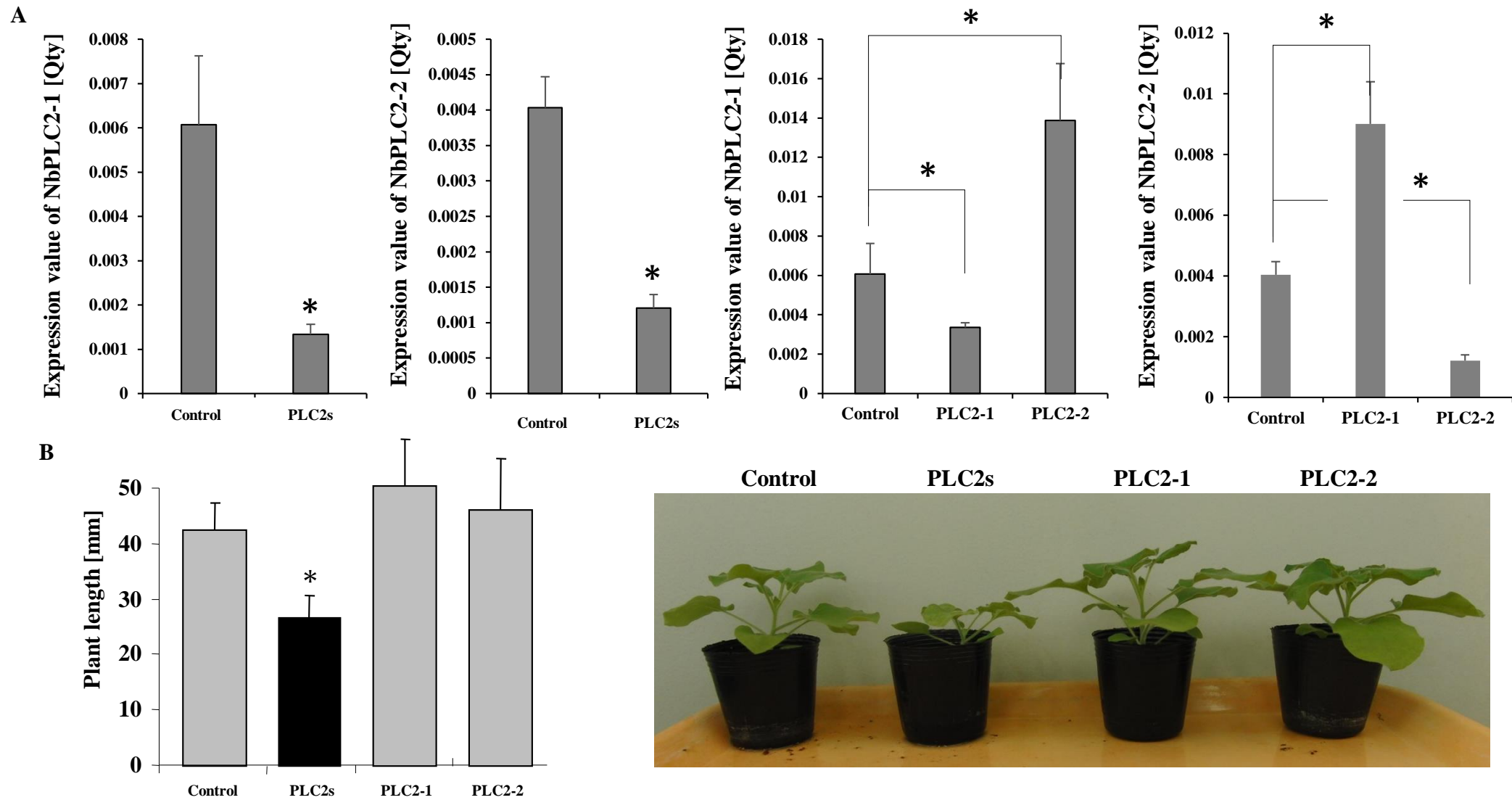


Figure S3 Phenotypic observation of *NbPLC2s*, *NbPLC2-1* and *NbPLC2-2*-silenced plants.

(A) Total RNA was isolated from control (Control), *NbPLC2s*-silenced (PLC2s), *NbPLC2-1*-silenced (PLC2-1) and *NbPLC2-2*-silenced (PLC2-2) leaves. Expression values were normalized against actin gene expression levels. Data represent the means and standard deviations from triplicate experiments. (B) Plant length was determined 3 weeks after inoculation with *Agrobacterium tumefaciens*. (C) Photograph was taken 3 weeks after inoculation with *Agrobacterium tumefaciens*. Values represent mean plant length ($n = 7$) with SD. Asterisks denote values significantly different from those of water-inoculated controls (*; $P < 0.05$, t -test).

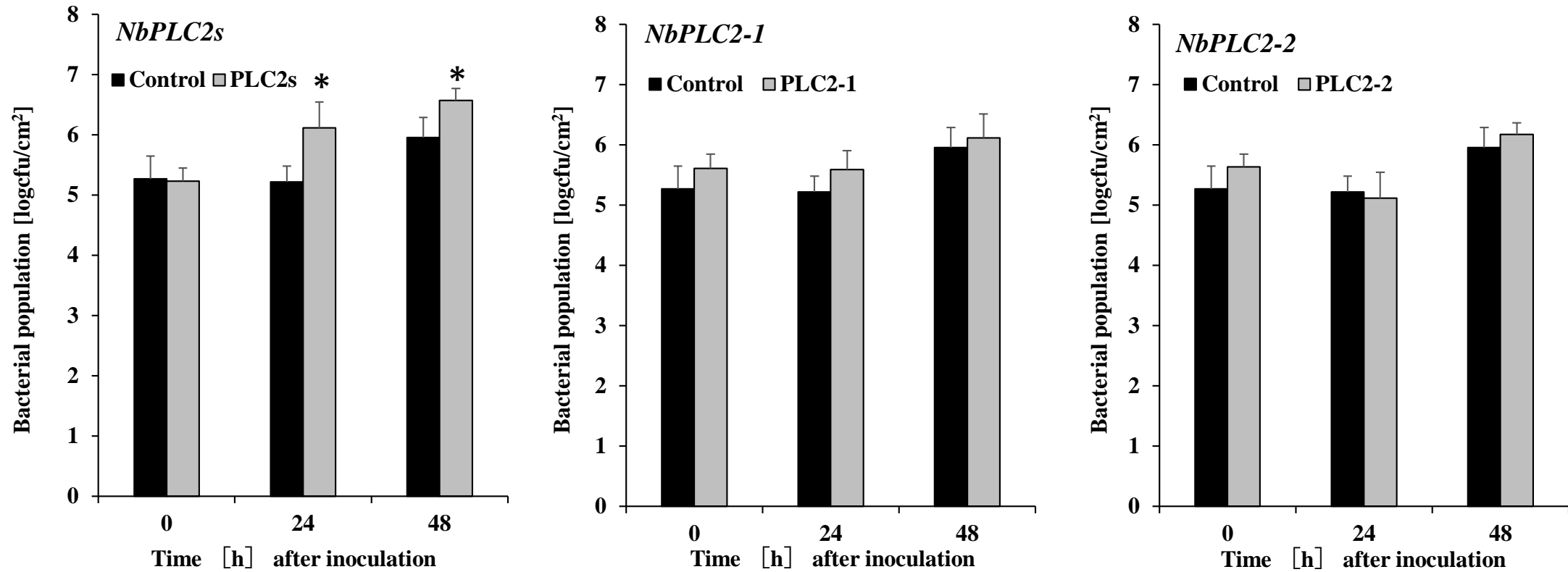


Figure S4 Response of *NbPLC2s*, *NbPLC2-1* and *NbPLC2-2*-silenced plants against compatible *R. solanacearum*.

Control and *NbPLC2s*, *NbPLC2-1* and *NbPLC2-2*-silenced plant leaves infiltrated with *R. solanacearum* OE1-1. Bacterial population of *R. solanacearum* OE1-1 was determined by plating at specified time points. Values are means of five replicate experiments with SD. Asterisks denote values significantly different from those of control plants (*; $P < 0.05$, *t*-test).

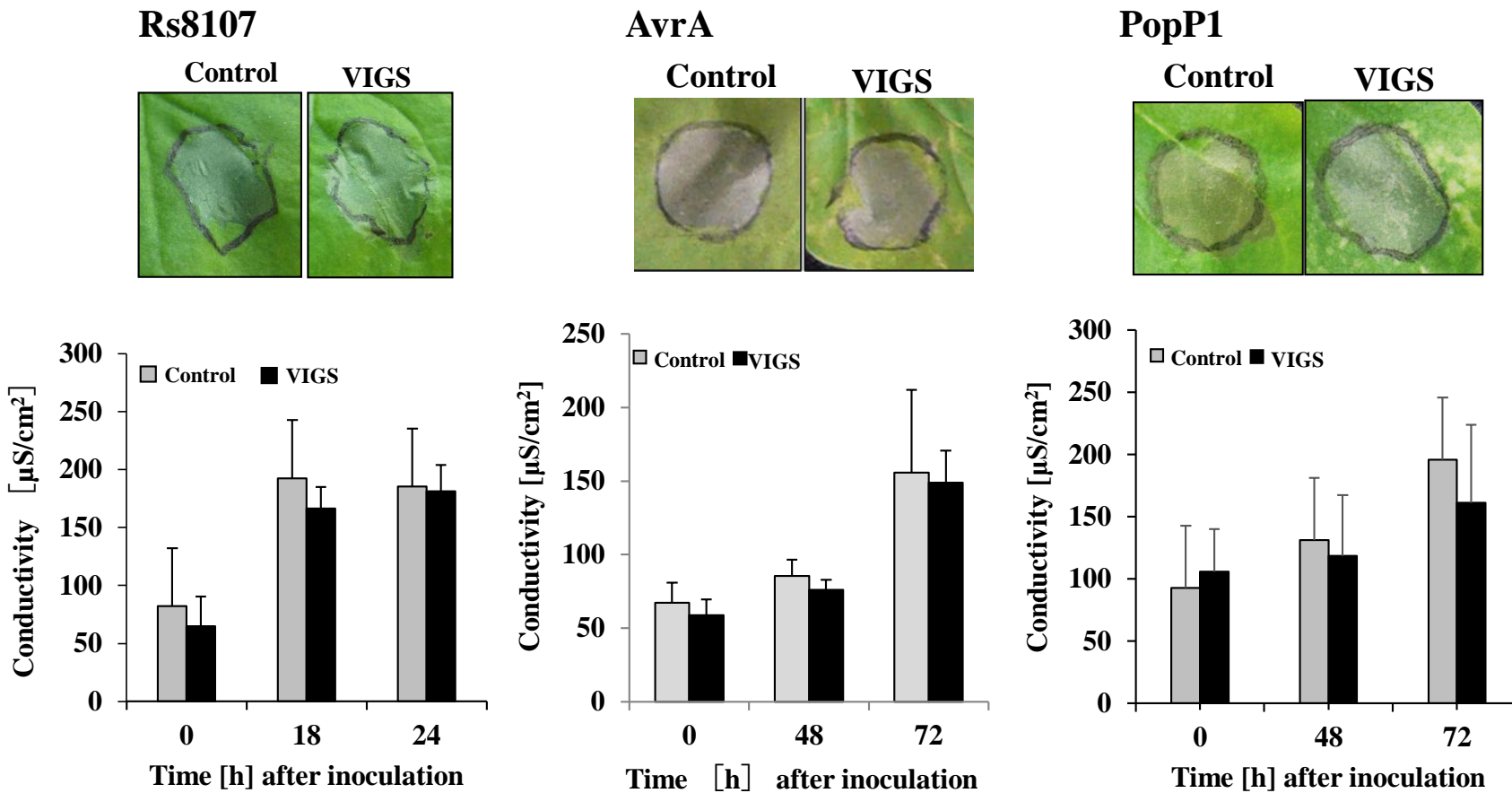


Figure S5 Effect of *NbPLC2s*-silencing on HR induction by effectors from incompatible *R. solanacearum*.

Control and *NbPLC2s*-silenced (VIGS) *N. benthamiana* plants were infiltrated with *R. solanacearum* 8107 (Rs8107), HR-inducible *Agrobacterium* harboring 35S-GUS (control GUS), 35S-AvrA (AvrA) and 35S-PopP1 (PopP1). (A) Pictures of *N. benthamiana* leaves taken 1 or 4 days after infiltration with *R. solanacearum* 8107 or *Agrobacterium*, respectively. (B) Cell death was determined by measurement of ion conductivity (leakage). Values represent mean with SD (n=5).

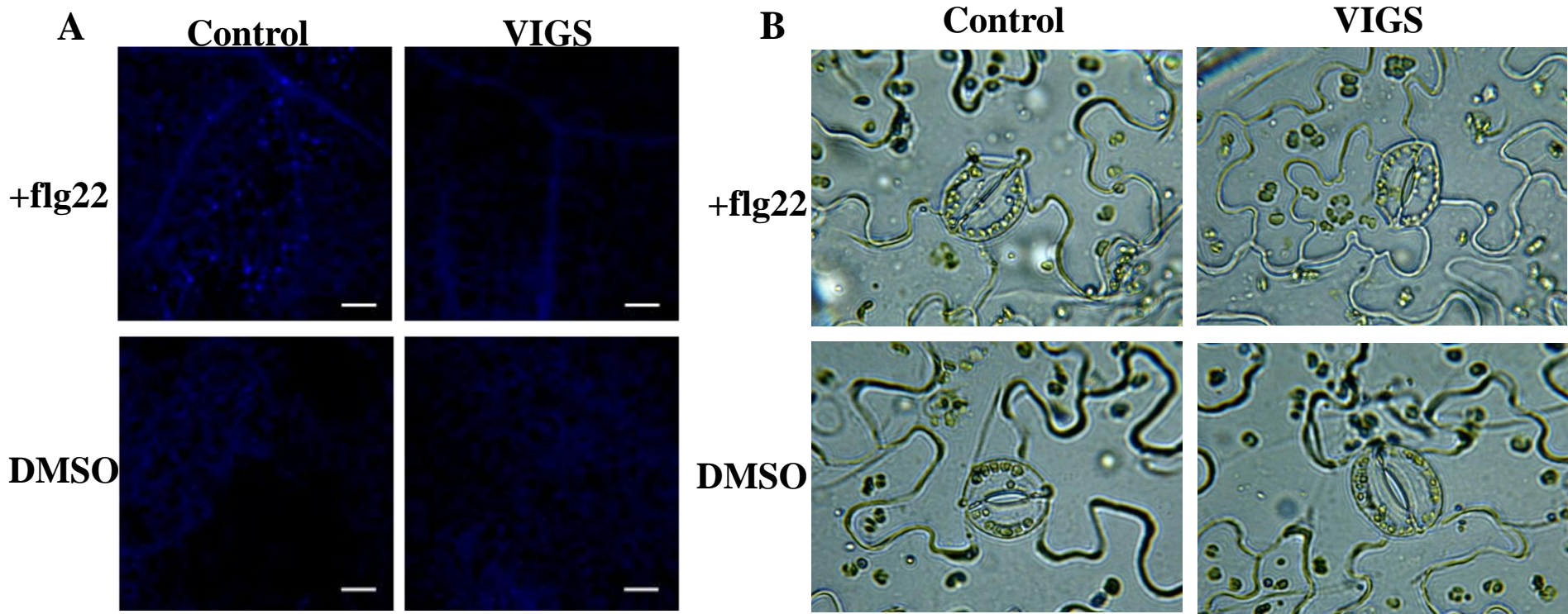


Figure S6 Characteristic of callose deposition and stomatal closure in *NbPLC2s*-silenced plant.

Effect of *NbPLC2s*-silencing on flg22-induced stomatal closure and callose deposition. Control and *NbPLC2s*-silenced (VIGS) leaves were treated with flg22 (+flg22) or DMSO (DMSO) solution. Observation of callose deposition (A) and stomatal closure (B) was carried out by the methods described in the materials and methods. Bar indicates 100 μ m.

Table S1 List of primers used in this study.

Primers	Sequence	Purpose
M4	5'-GTTTTCCCAGTCACGAC-3'	Sequencing
RV	5'-CAGGAAACAGCTATGGAC-3'	Sequencing
PLC2s VIGSS	5'- GTCGACCATCTAACTCCAGATCTTCAGGC-3'	For VIGS vector
PLC2s VIGSA	5'- GTCGACCTTTCTTTGAGGCATCTTTTCC-3'	For VIGS vector
PLC2-1 VIGSS	5'- GTCGACGGAAGGGAGAGAAGTACAG-3'	For VIGS vector
PLC2-1 VIGSA	5'- GTCGACATATTTCTCAACCATAACTCGAAC-3'	For VIGS vector
PLC2-2 VIGSS	5'- GTCGACGGAAGGGAGAGAAGTACAC-3'	For VIGS vector
PLC2-2 VIGSA	5'- GTCGACGGCAAAATATTTCTCAACTCG-3'	For VIGS vector
PLC2-1rtpF	5'- GGAGAAAAATGTAACCTTG-3'	qRT-PCR
PLC2-2rtpF	5'- GGAGAAAAATGTAACCTTA-3'	qRT-PCR
PLC2-1&2-2rtpR	5'- CCTGTGTATATGTAGTAATG-3'	qRT-PCR
NbAcre31rtp F	5'- AAGGTCCCCTCTTCGTCGGATCTTCG-3'	qRT-PCR
NbAcre31rtp R	5'- AAGAATTCGGCCATCGTGATCTTGGTC-3'	qRT-PCR
NbPti5rtp F	5'- CCTCCAAGTTTGAGCTCGGATAGT-3'	qRT-PCR
NbPti5rtp R	5'- CCAAGAAATTCTCCATGCACTCTGTC-3'	qRT-PCR
NbACTrtpF	5'-CGGAATCCACGAGACTACATAC-3'	qRT-PCR
NbACTrtpR	5'-GGGAAGCCAAGATAGAGC-3'	qRT-PCR
PR-4rtpF	5'-GGCCAAGATTCCTGTGGTAGAT-3'	qRT-PCR
PR-4rtpR	5'-CACTGTTGTTTGAGTTCCTGTTCCCT-3'	qRT-PCR

Table S2 List of plasmids used in this study.

Plasmid	Relevant characteristics	Reference
pMD20	Cloning vector pMD20, Amp ^r	Takara Bio
pMD-NbPLC2s	pMD20 carrying identical sequence of <i>NbPLC2-1</i> and <i>NbPLC2-2</i> fragment for VIGS	This study
pMD-NbPLC2-1	pMD20 carrying <i>NbPLC2-1</i> fragment for VIGS	This study
pMD-NbPLC2-2	pMD20 carrying full length <i>NbPLC2-2</i> fragment for VIGS	This study
pPVX201	pPVX binary vector, Km ^r	Baulcombe <i>et al.</i> , 1995
pPVXPLC2s	pPVX binary vector containing identical sequence of <i>NbPLC2-1</i> and <i>NbPLC2-2</i> fragment for VIGS	This study
pPVXPLC2-1	pPVX binary vector containing PLC2 -1 cDNA fragment for VIGS	This study
pPVXPLC2-2	pPVX binary vector containing PLC2 -2 cDNA fragment for VIGS	This study
P35S-GUS	pGreen binary vector carrying β -glucuronidase (GUS)	Katou <i>et al.</i> , 2003
P35S-AvrA	pMDC32-derived plasmid that contains the full-length <i>avrA</i>	Poueymiro <i>et al.</i> 2009
P35S-PopP1	pAM-PAT-P35S-GW-CFP-derived plasmid that contains the <i>popP1</i>	Poueymiro <i>et al.</i> 2009