

Table S1. Oligonucleotides used for genotyping and RT-PCR

Primer name	Symbol in figure 1	Sequences of primer	Uses	References
AtNOA1.1-LP	2	5'-GCACCTACACCACAGGCAAGC-3'	<i>noa1-2</i> genotyping <i>AtNOA1</i> RT-PCR	This work
AtNOA1.1-RP	3	5'-CCAATTGGCAATGTTGGTCG-3'	<i>noa1-2</i> genotyping <i>AtNOA1</i> RT-PCR	This work
CAPS NR1-F	4	5'-TACGACGACTCCTCAAGCGAC-3'	<i>nia1</i> genotyping	This work
CAPS NR1-R	5	5'-GGCTATAGATCCCGCATCGAC-3'	<i>nia1</i> genotyping	This work
NR2.1-LP	6	5'-ACGGCGTGGTTCGTTCTTACA-3'	<i>nia2</i> genotyping <i>NIA2</i> RT-PCR	This work
NR2.1-RP	7	5'-ACCTTCTTCGTCGGCGAGTTC-3'	<i>nia2</i> genotyping <i>NIA2</i> RT-PCR	This work
NR1.1-LP		5'-TCATAGCCGGAGAGGAGGCG-3'	<i>NIA1</i> RT-PCR	This work
NR1.1-RP		5'-CCATGAGGTTCCAGATGAGT-3'	<i>NIA1</i> RT-PCR	This work
SAIL LB3	1	5'-TAGCATCTGAATTTTCATAACCAATCTCGATACAC-3'	<i>noa1-2</i> genotyping	(a)
UBQ10-F		5'-GATCTTTGCCGAAAACAATTGGAGGATGGT-3'	RT-PCR normalization	(b)
UBQ10-R		5'-CGACTTGTCATTAGAAAGAAAGAGATAACAGG-3'	RT-PCR normalization	(b)
qRD29b-F		5'-CTTGGCACCACCGTTGGGACTA-3'	<i>RD29b</i> qRT-PCR	This work
qRD29b-R		5'-TCAGTTCCCAGAATCTTGAAC-3'	<i>RD29B</i> qRT-PCR	This work
qRAB18-F		5'-AAGAAGAACATGGCGTCTTACCA-3'	<i>RAB18</i> qRT-PCR	This work
qRAB18-R		5'-TGCTGCTGGATCGGGTTT-3'	<i>RAB18</i> qRT-PCR	This work
qACT2-F		5'-TTGTTCCAGCCCTCGTTTGT-3'	qRT-PCR normalization	(b)
qACT2-R		5'-TGTCTCGTGGATTCCAGCAG-3'	qRT-PCR normalization	(b)

(a) Mc Elver, J et al. (2001) *Genetics*, 159, 1751-1763(b) Castillo MC and León J. (2008) *Journal of Experimental Botany*, 59, 2171–2179

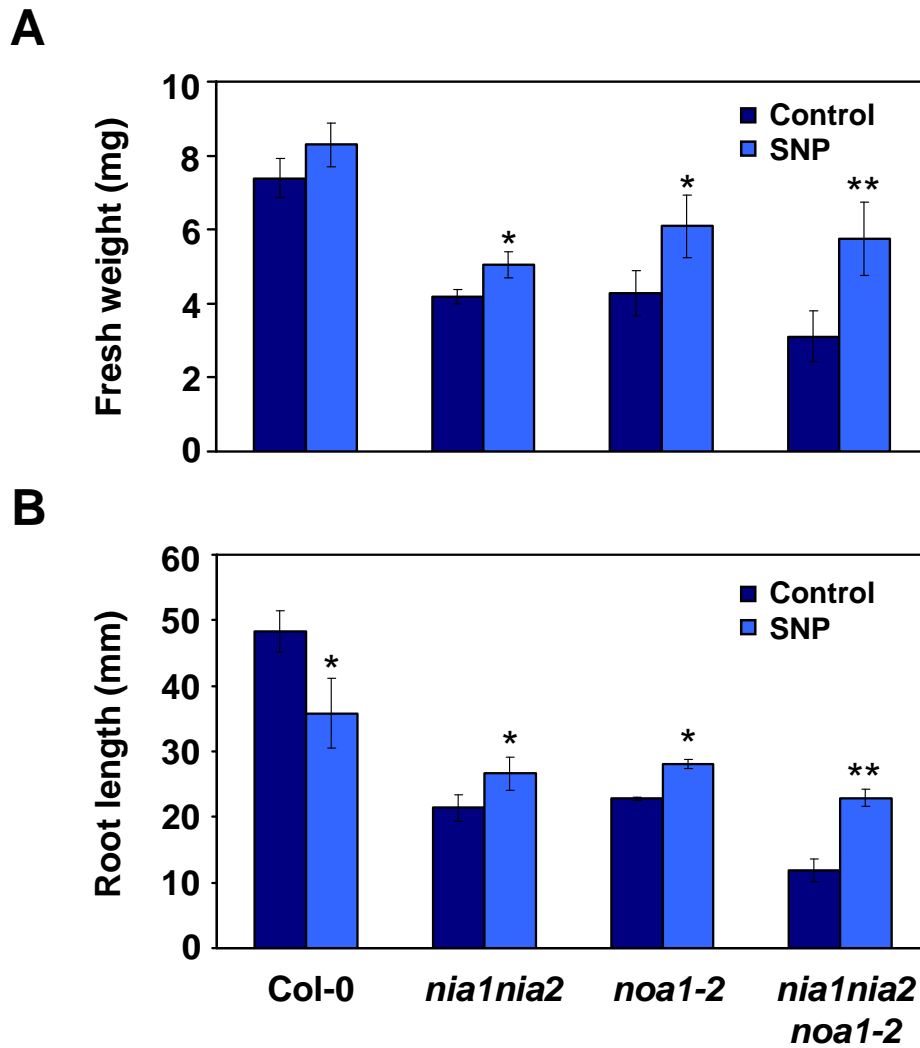


Figure S1. Rescue of vegetative developmental phenotypes of NO-deficient mutants by NO. (A) Fresh weight (mg) per seedling (n=12) of the indicated genotypes grown for 14 days in MS plates (Control) or 7 days in MS plates plus 7 days in MS plates supplemented with 100 μ M SNP (SNP) as indicated in Material and Methods. (B) Root length (mm) of the indicated genotypes grown in vertical plates for 14 days in MS (Control) or MS supplemented with 100 μ M SNP (SNP) as indicated in Material and Methods. Means \pm standard deviation are shown. Asterisks indicate statistical significance vs. every genotype control in each case (*p-value < 0.05; ** p-value < 0.01; Student's t-test).

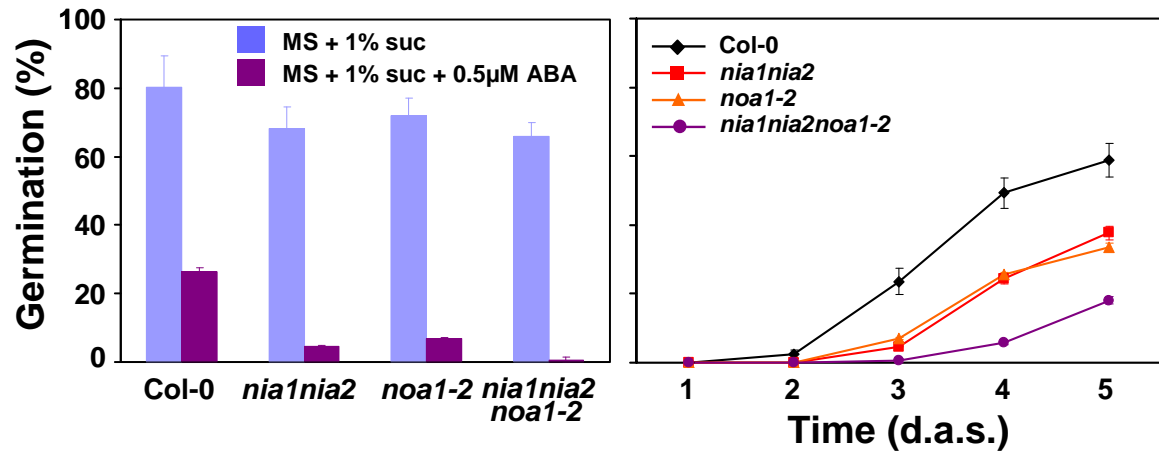


Figure S2. The NO-deficient mutant plants show hypersensitivity to ABA in germination assays. Left panel shows the percentage of germinated seeds (endosperm rupture) at day 3 after sown in the indicated media. Right panel shows the percentage of germinated seeds in MS media plus 0.5 μ M ABA scored daily to day 5. At least 200 seeds per genotype were sown, stratified for 3 days at 4 °C in each experiment. Values are mean \pm standard deviations. The experiment was repeated three times with similar results. MS plates: MS salts; 1 % sucrose; 0.8 % agar, pH=5.7.

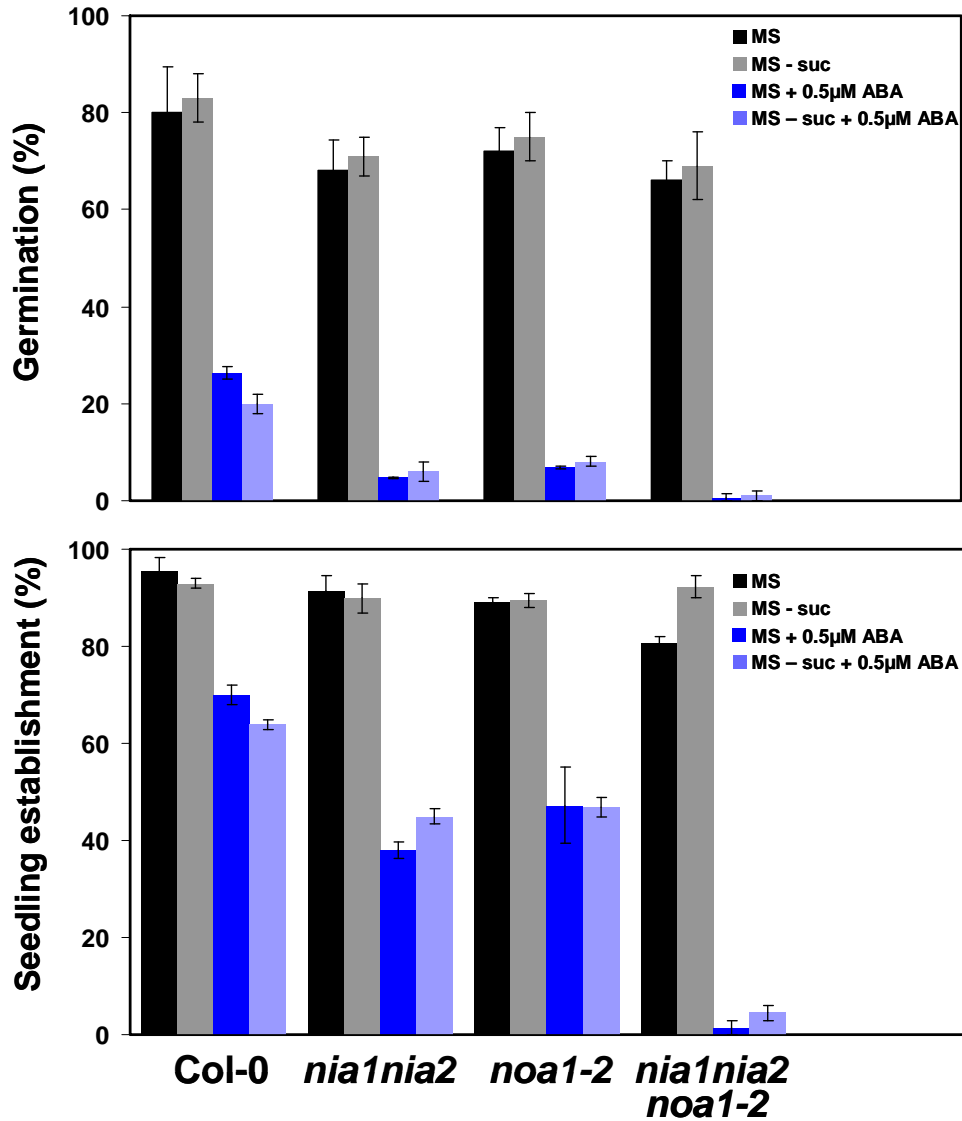


Figure S3. Sucrose content does not affect ABA sensitivity in NO-deficient mutant plants germination and establishment. Percentage of germination (endosperm rupture) at day 3 after sown (top panel) and percentage of seeds that develop green expanded cotyledons (seedling establishment) after 12 days after sown (bottom panel) of the indicated genotypes and media composition under LD conditions. MS: MS salts; 1 % sucrose; 0.8 % agar, pH=5.7; MS-suc: MS salts; 0.8 % agar, pH=5.7; MS + 0.5 µM ABA: MS salts; 0.5 µM ABA; 1 % sucrose; 0.8 % agar, pH=5.7; MS-suc + 0.5 µM ABA: MS salts; 0.5 µM ABA; 0.8 % agar, pH=5.7. Means ± standard deviations are shown. The experiment was repeated two times with similar results.

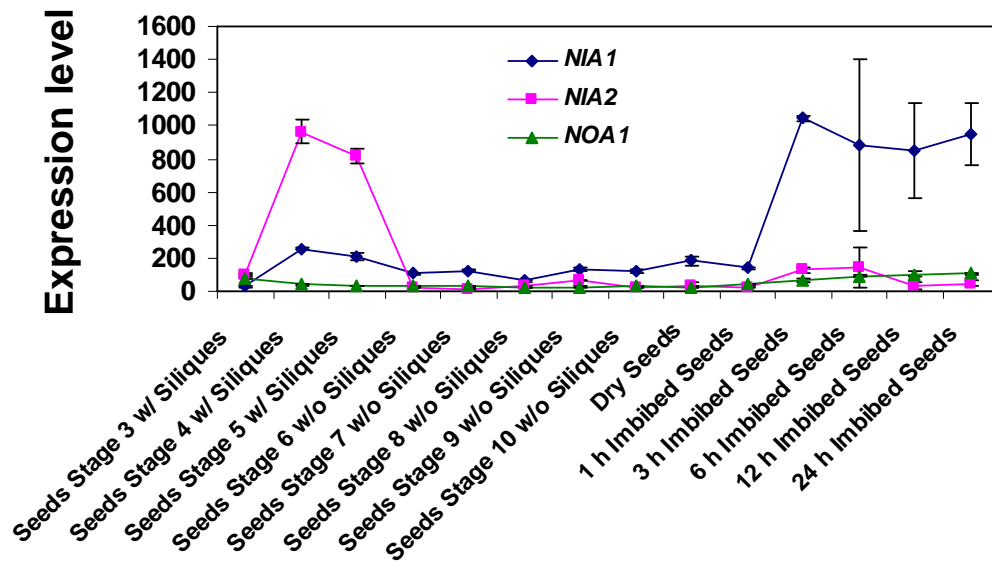
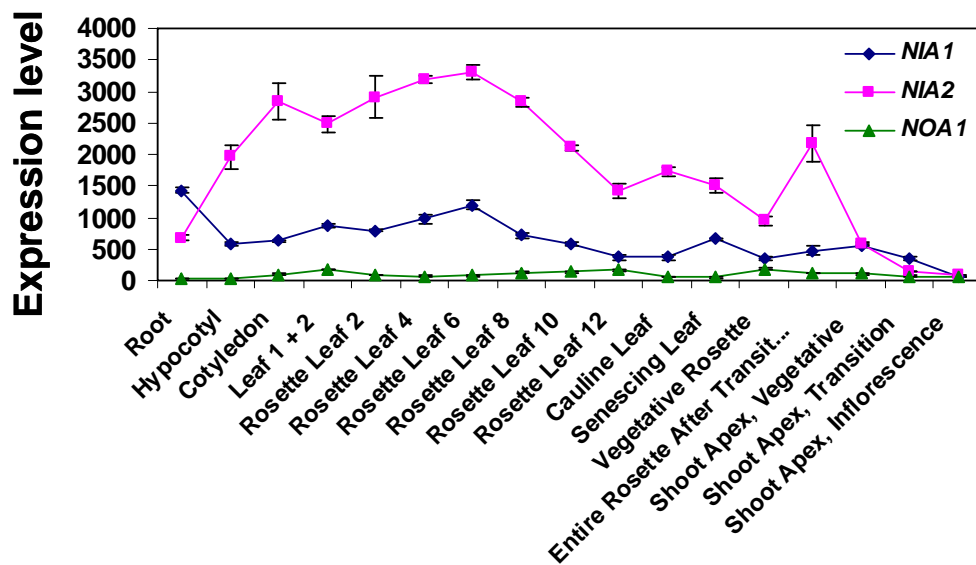
A**B**

Figure S4. *NIA1*, *NIA2* and *AtNOA1* expression levels in different tissues and developmental stages. (A) *NIA1*, *NIA2* and *AtNOA1* expression levels in seeds from seed stage 3 within siliques to 24h imbibed seeds. (B) *NIA1*, *NIA2* and *AtNOA1* expression levels in different tissues during development. Expression levels were taken from the public microarray database (BAR; <http://www.bar.utoronto.ca/>).

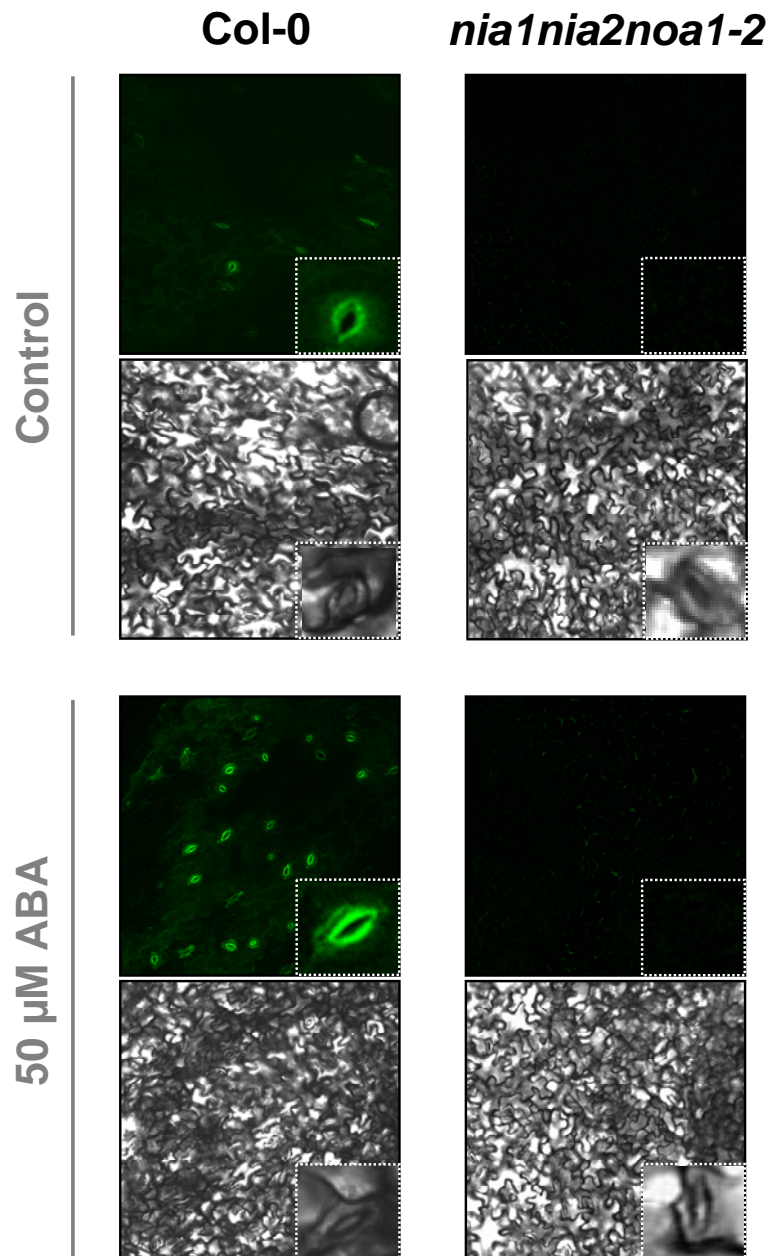


Figure S5. NO in guard cells of wild type and *nia1nia2noa1-2* stomata. Confocal microscope images of DAF-FM DA stained leaves treated or not with 50 μ M ABA for 30 min. Images were recorded with the same settings for the microscope and the laser source. Insets represents a magnification of the same stomata observed under fluorescence (up) or bright field (down).