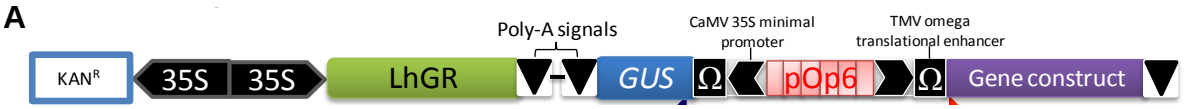

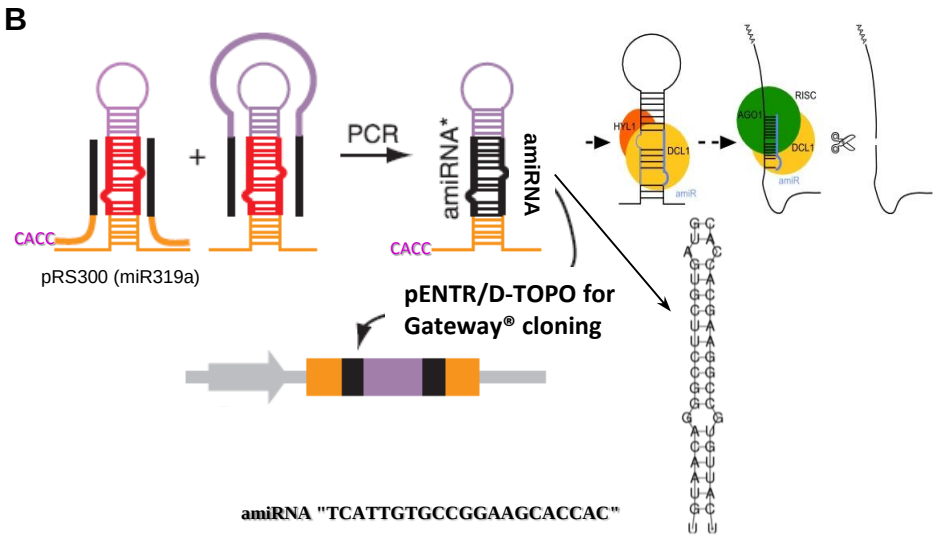

C

Sequence of amiREXPA10,1,5,3:

CACCCAAGGCGATTAAGTTGGGTAACGCCAGGGTTTTCCCAGTCACGACGTTGTAAAACGACGGCCAGTGAATTGTAATACGACTCACTATAGGGCGA
ATTGGGTACCGGGCCCCCCCTCGAGGTCGACGGTATCGATAAGCTTGATATCGAATTCCTGCAGCCCcaaacacacgctcggacgcatattacacatgttcatacactt
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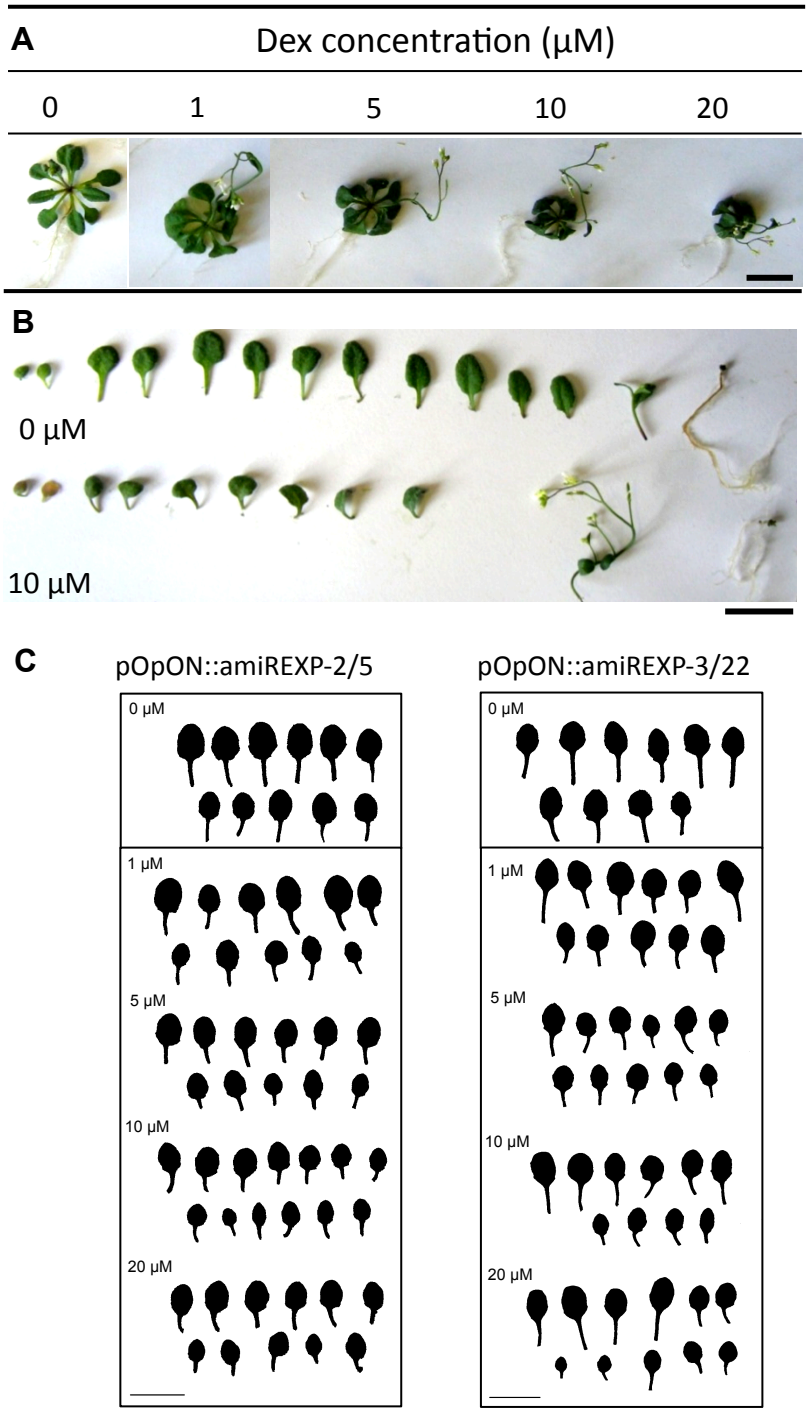
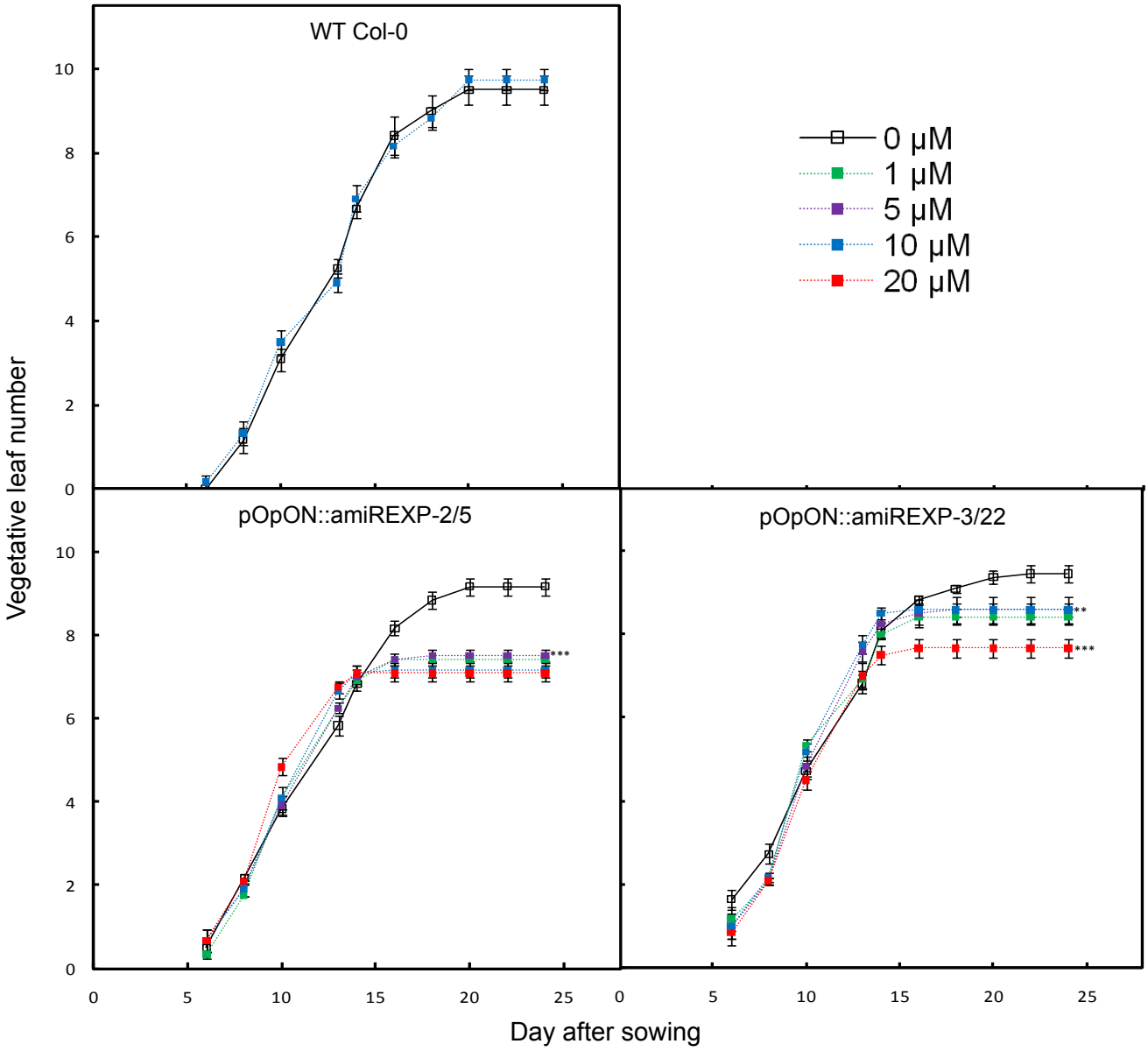
TAGTTCTAGAGCGGCCGCCACCGCGGTGGAGCTCCAGCTTTTGTTCCCTTTAGTGAGGGTTAATTTCGAGCTTGGCGTAATCATGGTCATAGCTGTTTCC
TGTGTGAAATTGTTATCCGC

T7 SP6 oligoA oligoB miRNA miRNA*

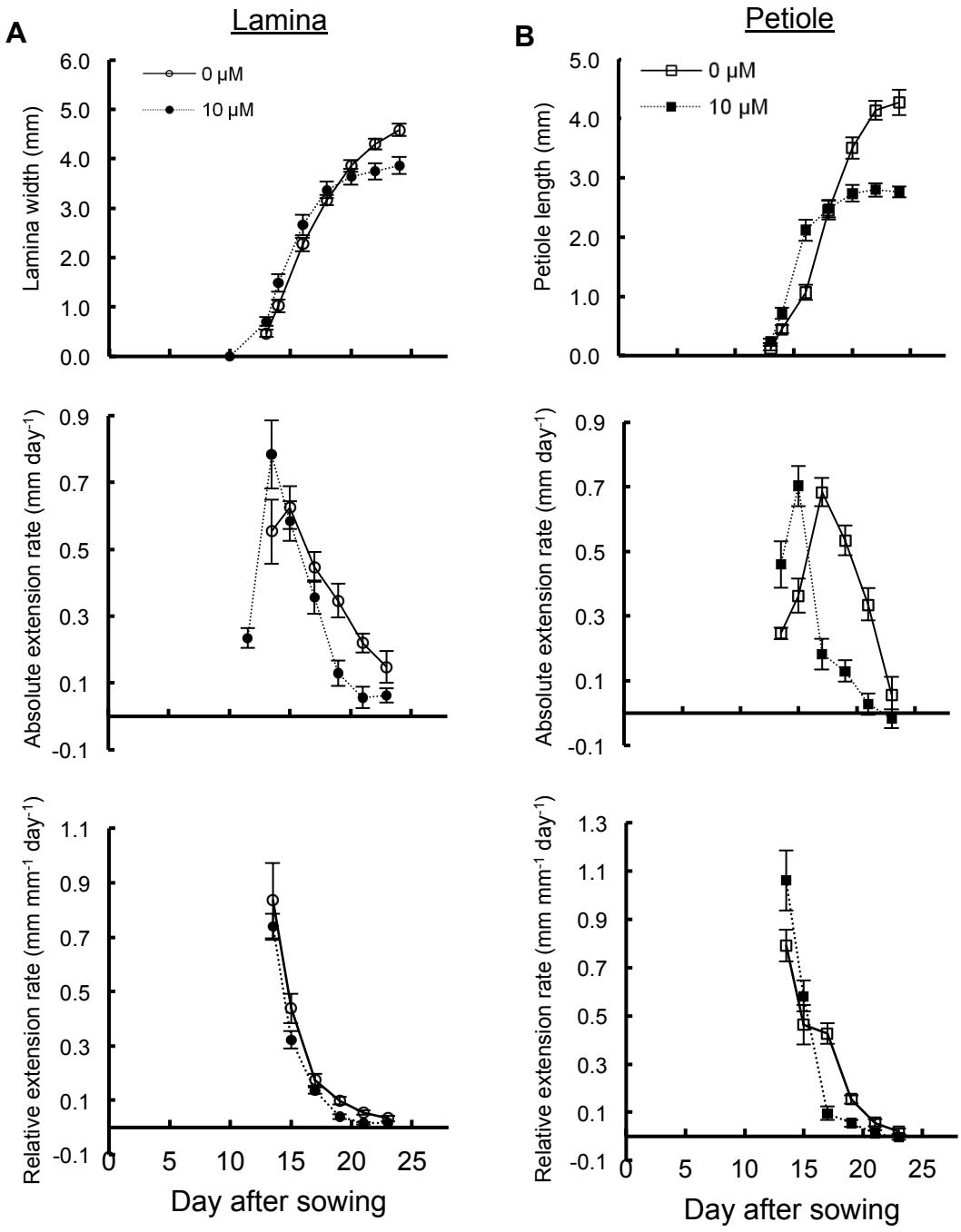
Supplemental Figure 1. Constructs for inducible suppression of expansin gene expression. **A**, Schematic diagram of the pOpON^{Kan} two-component inducible system with kanamycin resistance selectable marker. The dexamethasone-inducible pOpON2.1 transactivating system contains two transcription units. The first unit employs a constitutive CaMV 35S promoter to express a Dex-responsive chimaeric transcription factor (LhGR). LhGR is an artificial fusion between a high-affinity DNA-binding mutant of *lac* repressor, transcription-activation domain-II of GAL4 from *Saccharomyces cerevisiae* and ligand-binding domain of a rat glucocorticoid receptor (GR LBD). The second unit consists of six copies of the multimerised *lac* operators (transcription factor-binding site pOp6) linked to a truncated 35S promoter, which forms the bidirectional promoter used to express the construct of interest and expression marker *b*-glucuronidase (*GUS*) gene. Each construct ends with poly-A signal that serves as transcription terminator. Circles representing dexamethasone (Dex) which binds and activates the constitutively expressed LhGR, in turn results in the transcription of both the gene construct and *GUS* **B**, Artificial miRNA (amiR) approach for expansin gene silencing with schematic representation of amiRNA work flow adapted from (Ossowski et al., 2008). First, the customised amiRNA was generated through overlapping PCR with modified primers for Gateway-compatible cloning into the pOpON^{Kan} two-component inducible system. This allows targeted transcript cleavage-mediated silencing showing its predicted structure (Source: http://wmd3.weigelworld.org/cgi-bin/webapp.cgi). **C**, Sequence of the designed amiREXP for the down-regulation of *EXPA10,1, 5, 3* (in descending order of target specificity) highlighting functional sites.



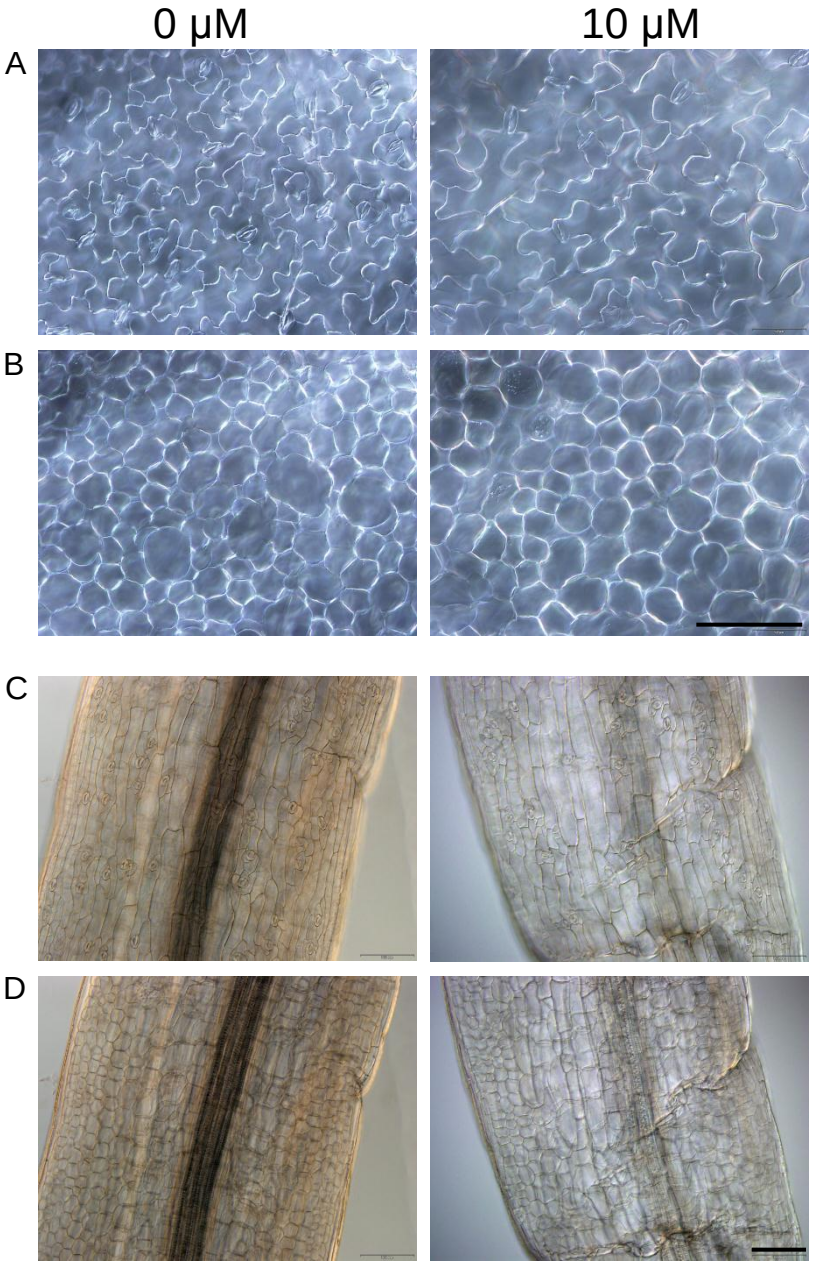
Supplemental Figure 2. A, Phenotype of induced pOpON::amiREXP (line 2/5) plants grown on medium supplemented with different concentrations of Dex or control medium 24 day after sowing (DAS), with **B**, representative non-induced (0 µM) and induced (10 µM) plants dissected for comparison of individual cotyledons, vegetative leaves, inflorescence and root. **C**, Silhouettes of leaf 6 from two independent lines of pOpON:amiREXP as described above, used for morphometric comparison (Fig. 5). Scale bars = 10 mm.



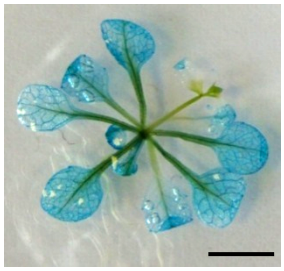
Supplemental Figure 3. Changes of vegetative leaf number over time (6 to 24 day after sowing) Col-0 wild-type, and pOpON::amiREXP line 2/5 and 3/22 plants under different Dex concentrations. Number of visible leaves (>1 mm width) from 12 individual plants of each line (mean ± SE), grown on half-strength MS agar medium supplemented with different concentrations of Dex or DMSO 0.1% (0 µM), were counted. Mann-Whitney tests or One-way ANOVA, Tukey tests compared to 0 µM control *p<0.05, **p<0.01, ***p<0.001, asterisks were annotated only for the first instances when the difference become significant or when the significance level changes and left out subsequently for clarity.



Supplemental Figure 4. Growth suppression following induction of independent line (2/5) pOpON::amiREXP plants. **A**, Lamina width, absolute lamina extension rate, and relative lamina extension rate of leaf 6 petioles from of pOpON::amiREXP plants grown continually either with or without 10 µM Dex. **B**, Petiole length, absolute petiole extension rate, and relative petiole extension rate as per A. Values are means ± SE (n = 12).



Supplemental Figure 5. Histological comparison of middle regions of leaf 6 lamina and petiole showing representative images from pOpON::amiREXP transformant plants grown on 0.5x MS agar medium supplemented with DMSO 0.1% or 10 µM Dex. **A**, Lamina adaxial epidermis. **B**, Lamina mesophyll. **C**, Petiole adaxial epidermis. **D**, Petiole mesophyll. Scale bars = 100 µm.



Supplemental Figure 6. GUS histochemistry of a pOpON::amiREXP plant 8 days after induction at 12 DAS with 10μM Dex at the shoot apex. Analysis was performed at 20 DAS with blue coloration indicating the extent of transcriptional induction through all visible leaves. Scale bar = 5 mm.

Supplemental Table 1. Primer sequences for artificial miRNA cloning.

Oligo name	Sequence (5'→ 3')
OligoA	CACCCAAGGCGATTAAGTTGGGTAAC
OligoB	GCGGATAACAATTTCACACAGGAAACAG
Ami10,1,5,3_I	gaTCATTGTGCCGGAAGCACCACtctctcttttgtattcc
Ami10,1,5,3_II	gaGTGGTGCTTCCGGCACAATGAtcaaagagaatcaatga
Ami10,1,5,3_III	gaGTAGTGCTTCCGGGACAATGTtcacaggtcgtgatatg
Ami10,1,5,3_IV	gaACATTGTCCCGGAAGCACTACtctacatatatattcct

Notes: Oligonucleotides A and B are based on the template plasmid sequence (pRS300) which are located outside of the multiple cloning site. Primer I contains the amiRNA in sense orientation, primer II its reverse complement, primer III the amiRNA* sequence in sense and primer IV the amiRNA* sequence in antisense orientation
The capital letters in oligo I to IV correspond to the desired miRNA-miRNA* region. The addition of CACC to OligoA allows pENTR/ D-TOPO cloning.

Supplemental Table 2. Sequences of primer pairs used in *EXPA* RT-PCR transcript analysis.

Gene	Accession no.	Sequence 5' → 3'
AtEXPA1	At1g69530	ACGTAGCTGCGCACCTGTGAA
		ACGCCTTACCGAACAACGCAG
AtEXPA2	At5g05290	AAATTGTCCGCCTTCAAAAGTCT
		CTTTGCCTTAGCTAATGATAATGG
AtEXPA3	At2g37640	GACTCGAAAGTTTTTGCCGGAG
		TTTGCTCAGCCGAGTGACGAC
AtEXPA4	At2g39700	GCTCGAAGGCGCTCTTGTTC
		TTGCGGTGATGAGGATCGAA
AtEXPA5	At3g29030	ATACCGAAACTGCCCTCCGGT
		CATTGTGGTCACTGCCACTAAC
AtEXPA6	At2g28950	GACTCTGAAGTTCTTTCCCATGA
		ACTTTGCTCAGCCTAGTGACAAT
AtEXPA7	At1g12560	ACGGAAATTAGCGGTGCTCTTG
		TTGGTACCAAGACTCCAACGCT
AtEXPA8	At2g40610	GAACTGACCACCTTGGTAGGTT
		CCCTGGCCTCTCCAACGATAA
AtEXPA9	At5g02260	GACGCGGAAGTTCTTGCCGG
		TCCTAACTTTAATCAAGCTAGCGA
AtEXPA10	At1g26770	CTTCTGCCCGCCAAATAACG
		GTCCACCGGCAAAAGTCTGG
AtEXPA11	At1g20190	GACTCTGAAGTTCTTTCCCATGA
		ACTTTGCTCAGCCTAGTGACAAT
AtEXPA12	At3g15370	ACGGAAATTAGCGGTGCTCTTG
		TTGGTACCAAGACTCCAACGCT
AtEXPA13	At3g03220	GAACTGACCACCTTGGTAGGTT
		CCCTGGCCTCTCCAACGATAA
AtEXPA14	At5g56320	GACGCGGAAGTTCTTGCCGG
		TCCTAACTTTAATCAAGCTAGCGA
AtEXPA15	At2g03090	CGGCCGTCAATTCCGTTAAA
		AGGCGGGTGGTGAAATTCCT
AtEXPA16	At3g55500	GTGGAAACGATGCTTCGGGA
		CTCCGGCCACGTTCGTAATC
AtEXPA17	At4g01630	GAAGTTAACGTTGCTCTTGAAGC
		AACTTTGCTCAGGCAAGCGACA
AtEXPA18	At1g62980	GCATGCGGTCAATGTTTCCA
		AGAGGTGAGCCGGAACGAGA
AtEXPA20	At4g38210	AGGAGTGGAACTGCTTTCCTTG
		ATTCGGGACTCTCCTCCGATTA
AtEXPA21	At5g39260	AAAGTTAGTCTTTCCATCAAAAGTC
		TTTATGTCCACCAGGATCCGCT
AtEXPA22	At5g39270	AAAGTTAATCCTACCATCAAAAGTC
		AATTACAGCAAAACCACAGACCTT
AtEXPA23	At5g39280	AAAGTTAATCTTTCCATCAAAAGTCT
		ATTACAGCAAAACCGAAGGCGTT
AtEXPA24	At5g39310	AAAATTAACCTTTCCATCAAAACTCT
		GATTTTACCAAGCCCAATGACAAT
AtEXPA25	At5g39300	AAAGTTAATCTTTCCATCAAAAGTCT
		ATTACTCCAAAACCGAAGGCGTT
AtEXPA26	At5g39290	AAAAGTTAATCTTACCATCAAAAGTC
		AATTACAGCAAAACCACAGACCTT

Supplemental Table 3. Sequences of primers used for RT-qPCR showing the results from standard graphs.

Gene	Oligo	Sequence (5'→ 3')	Amplicon		Standard graph		
			Size (bp)	Tm	Slope	R^2	Efficiency
AtEXPA1	FP	CTTACCGAAGAGTGCCGTGCGT	102	78	-2.95	0.99	118
	RP	CGGCTCCTCCGACGTTAGTG					
AtEXPA3	FP	CCCGTCTCCTATCGCAGGGTA	106	78	-2.92	0.99	120
	RP	CGCCGGCAACGTTAGTTACC					
AtEXPA5	FP	ATGGTACCCTCCCAACCACCA	109	77	-2.77	0.99	130
	RP	CGAGCTCCGAACCCTTCTATACATAAC					
AtEXPA8	FP	GGCTCTATTCAACAACGGACTCA	79	68	-3.09	0.99	110
	RP	CGGGTCATCGTTACACTTCATCT					
AtEXPA10	FP	AACCCAACAACATAATTTCACATA	82	73	-2.95	0.98	126
	RP	TGTTAACAACTTGGGACTTGG					
AtEXPA13	FP	CCGGTGCAGTATCGAAGGATCAAC	70	78	Not determined[a]		
	RP	TGCCTCCACCATCGACTGTAAAC					
AtEXPA15	FP	CTTCTCATACACGGAACCCTTCTG	77	76	-2.78	0.90	129
	RP	GCATTGCTCAGTACAAAGCTGGTG					
UBC21	FP	CTCTTAACTGCGACTCAGGGAATC	67	75	-3.49	1	93
(At5g25760)	RP	TGTGCCATTGAATTGAACCCTCTC					
ACT2	FP	CTTCCGCTCTTTCTTTCCAAGCTC	76	75	-3.31	0.99	100
(At3g18780)	RP	ACCATTGTCACACACGATTGGTTG					

[a]A primer efficiency value of 100 was used whenever the exact efficiencies of primer pairs were not determined