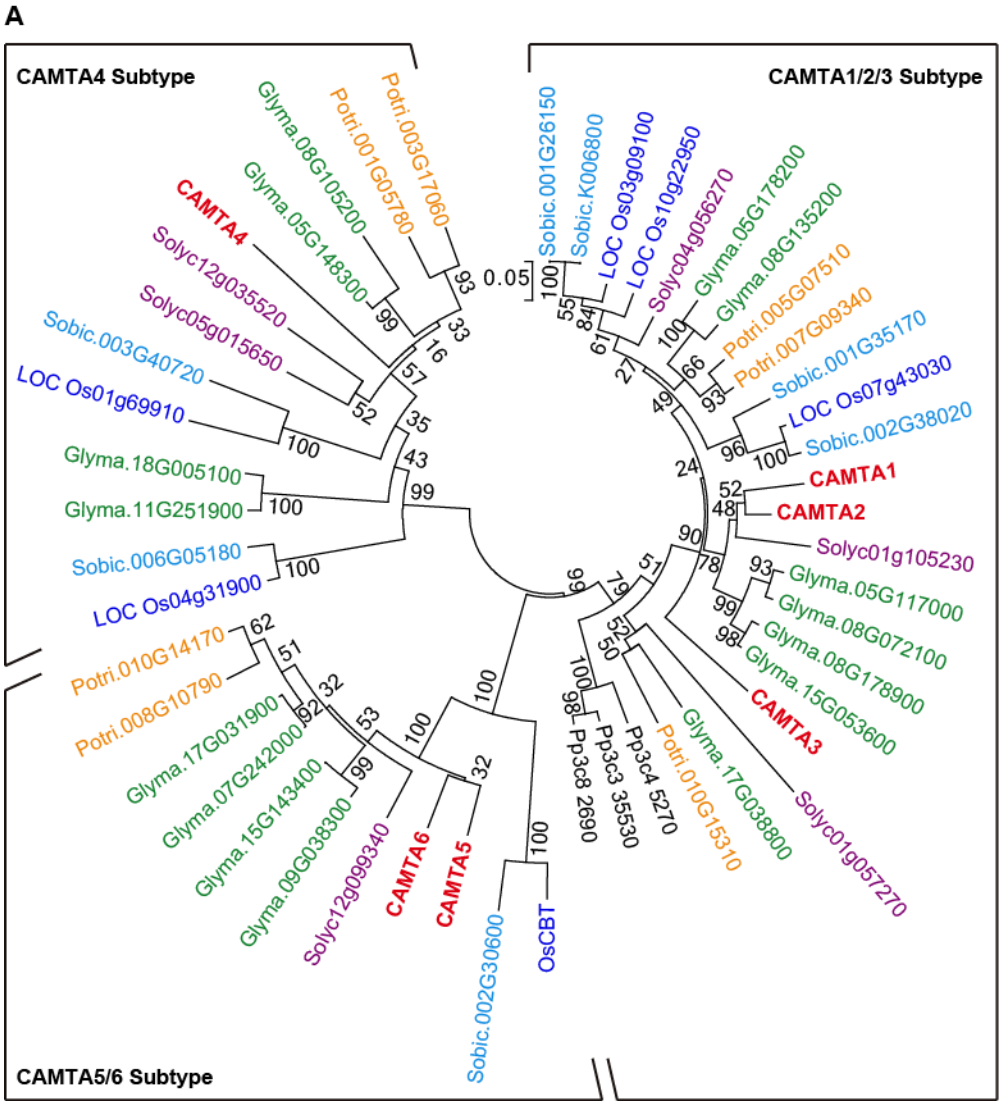
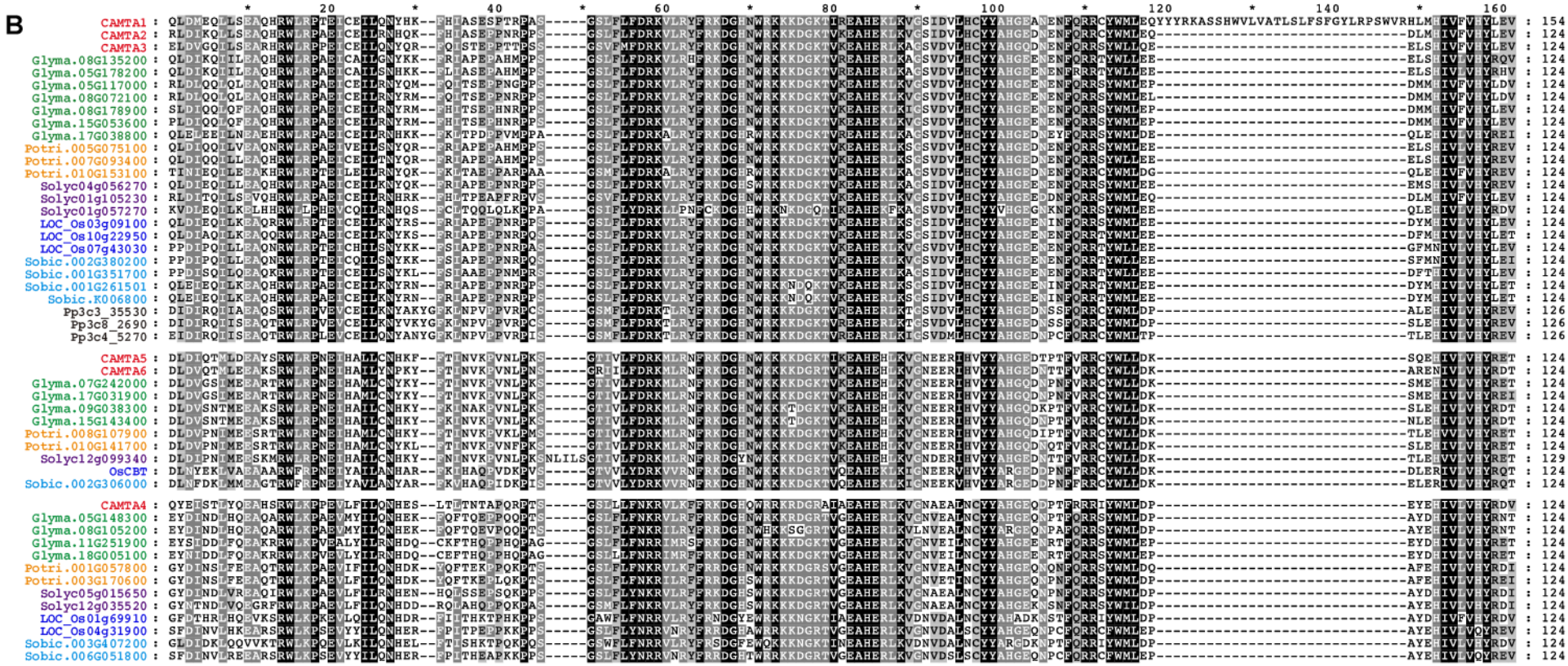
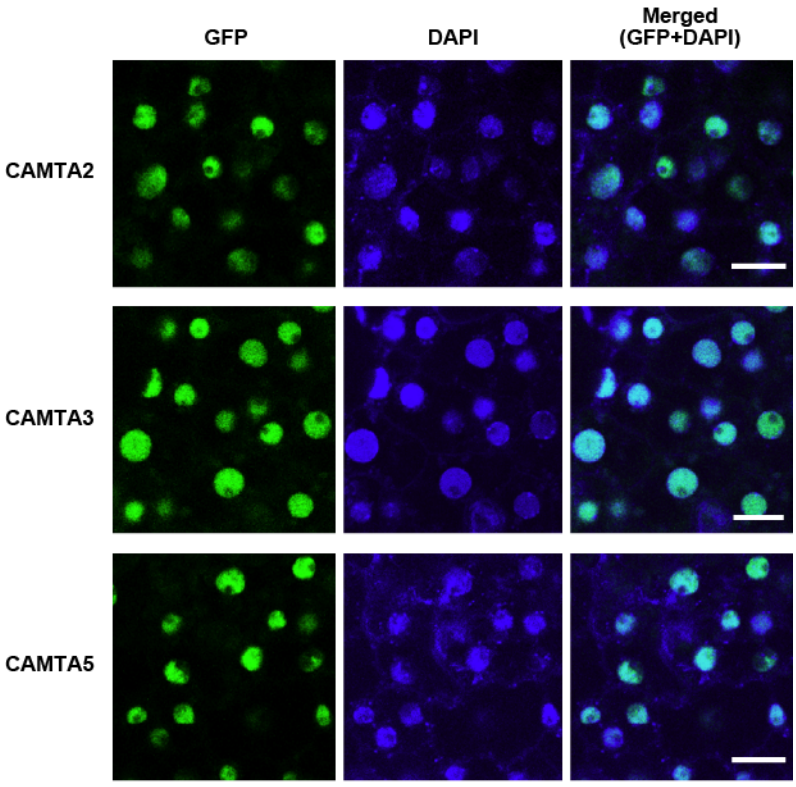


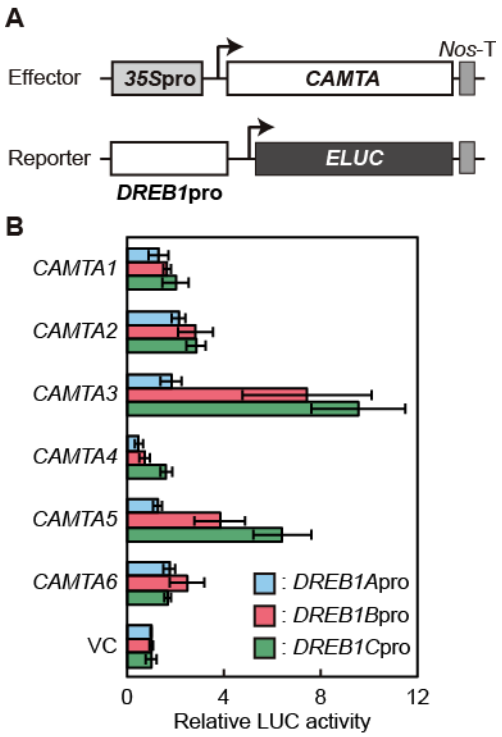
Supplemental Figure 1. Phylogenetic tree of CAMTA family proteins.
(Supports Figure 1.)
(A) Phylogenetic tree constructed according to the CG1 DNA-binding domains of the CAMTA transcription factors in *Arabidopsis thaliana* (red), *Glycine max* (green), *Solanum lycopersicum* (purple), *Populus trichocarpa* (orange), *Oryza sativa* (blue), *Sorghum bicolor* (aqua) and *Physcomitrella patens* (black). A total of 45 candidate peptide sequences were identified in the Phytozome v11.0 genomic database (http://www.phytozome.net/).



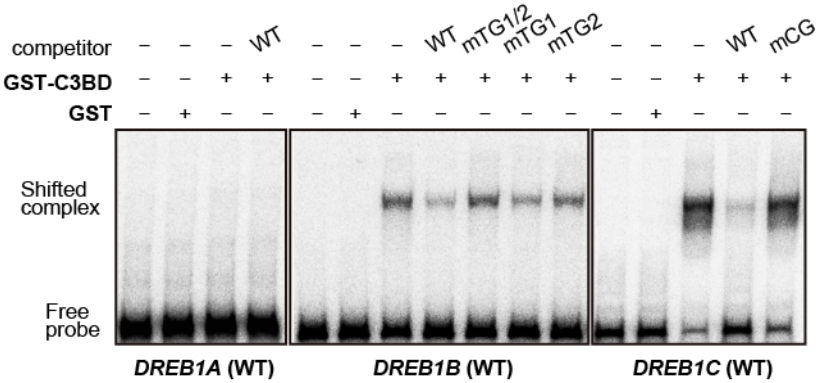
Supplemental Figure 1. (continued)
(B) Alignment of the CG1 DNA-binding domains used to construct the phylogenetic tree shown in (A).



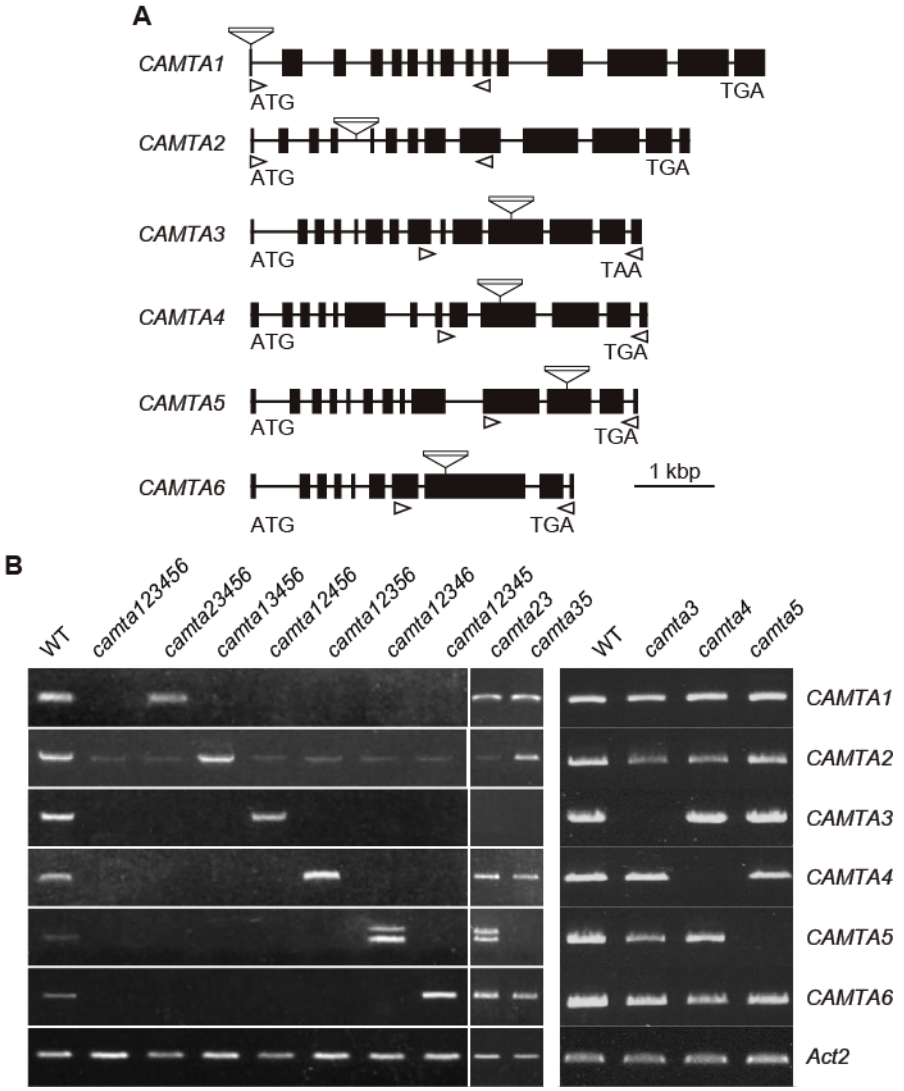
Supplemental Figure 2. Subcellular localization of CAMTA proteins in transgenic plants.
(Supports Figure 1.)
Expression of *CAMTA-GFP* fusion genes driven by the CaMV *35S* promoter was observed in 1-week-old seedlings. Rosette leaves of *CAMTA2*-, *CAMTA3*- and *CAMTA5-GFP* plants are shown. GFP fluorescence, DAPI fluorescence and differential interference contrast (DIC) images are presented. Bars = 10 μm.



Supplemental Figure 3. Transactivation analyses of the CAMTA family proteins using each 1-kb *DREB1* promoter.
(Supports Figure 2.)
(A) Schematic diagrams of the effector and reporter constructs used in transactivation analyses. The effector constructs contained the CaMV *35S* promoter fused to the CAMTA1-6 coding sequences. *Nos*-T indicates the polyadenylation signal of the nopaline synthase gene. The reporter constructs contained the *emerald LUC* gene fused to a 1-kb fragment of the *DREB1A, DREB1B* or *DREB1C* promoter.
(B) Transactivation activities of the CAMTA family proteins in Arabidopsis mesophyll protoplasts. The relative activity indicates the fold change in expression compared with the expression in the vector control. The error bars indicate the standard deviation of three biological replicates.



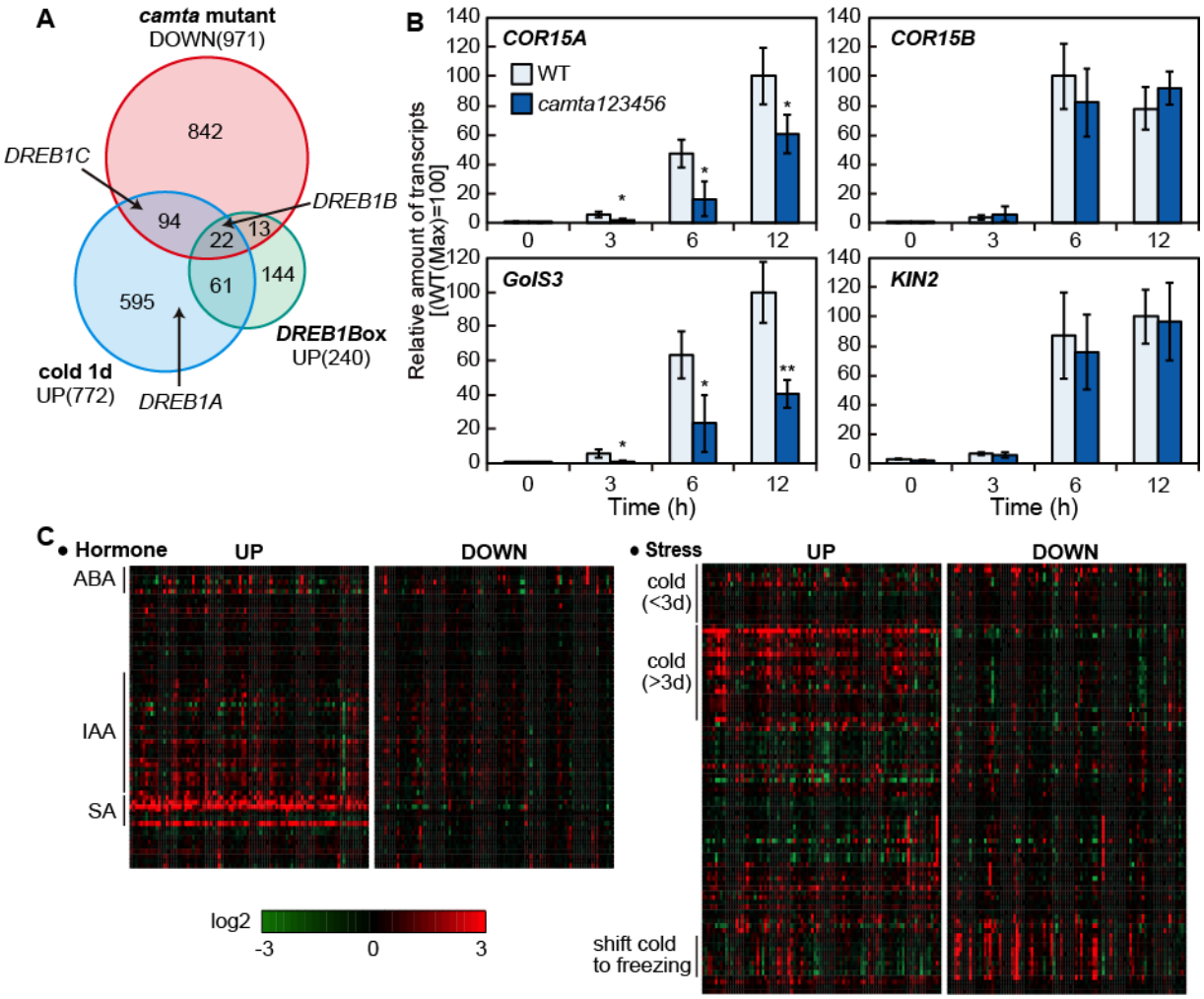
Supplemental Figure 4. Electrophoresis mobility shift assay of binding of the recombinant CAMTA3BD protein to the *DREB1* promoters.
(Supports Figure 2.)
Radioactive probes of *DREB1A*, *DREB1B* and *DREB1C* were incubated with or without a 1,000-fold excess of competitors in the presence of the recombinant CAMTA3BD protein (GST-C3BD). The DNA fragments of probes and competitors were the same as those in Figure 2C.



Supplemental Figure 5. Construction of mutants of the *CAMTA* family genes. (Supports Figure 3.)

(A) Schematic diagrams of the *CAMTA* genes. The exons (boxes) and introns (line) are indicated. The positions of T-DNA insertions are shown. The arrowheads indicate the RT-PCR primer positions.

(B) Expression of the *CAMTA* genes in the WT and mutant plants used in this study, as determined by RT-PCR.

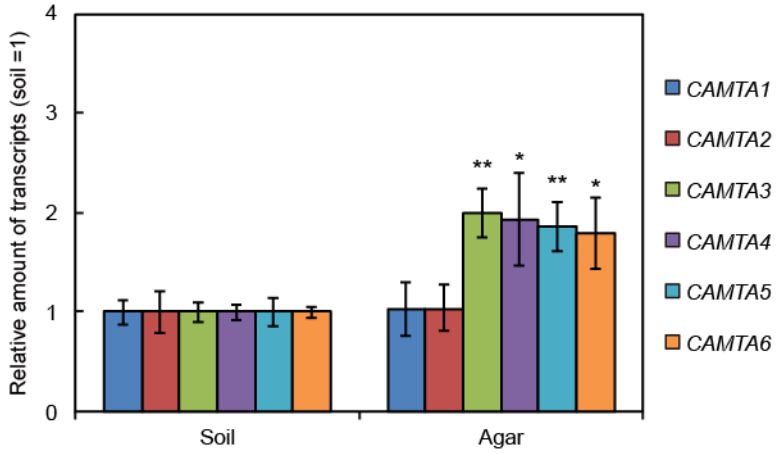


Supplemental Figure 6. Transcriptome analysis of sextuple mutants of the *CAMTA* family genes grown in soil pots.
(Supports Figure 3.)

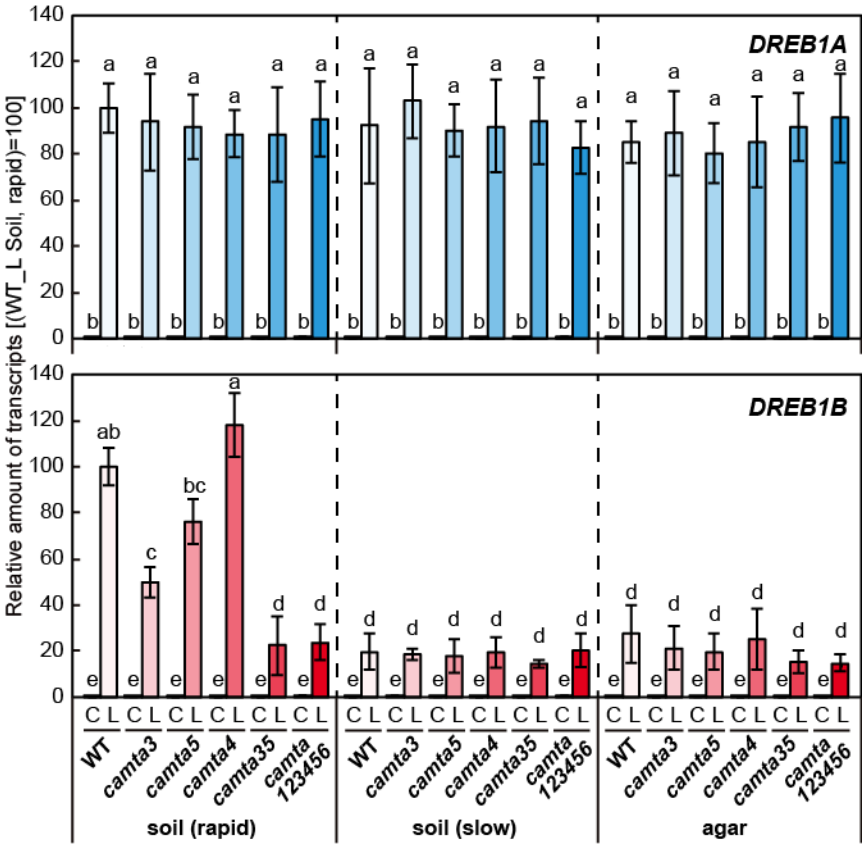
(A) Venn diagram comparing the down-regulated genes in the *camta* mutants with cold-inducible and DREB1B-downstream genes. The total number of genes in each group is shown in parentheses.

(B) Transcript levels of the DREB1-downstream genes whose expression was down-regulated or unchanged in the *camta* sextuple mutant under cold stress conditions. Three-week-old plants grown in soil pots under a 12-h light/12-h dark cycle at 22°C were immediately cooled to 4°C beginning at ZT2. The transcript level of each gene in the plants treated for the indicated duration (0, 3, 6, or 12 h) was measured by RT-qPCR. The values represent the average of three replicates, and the error bars indicate the SD. The maximum expression level of each gene in the WT plants was set to 100. The asterisks indicate significant differences (*p < 0.05 and **p < 0.01 according to Student's t-test) in the expression of each gene compared with that in the WT plants at each time point in three biological replicates using the plants sampled at different times.

(C) Responses of the up- and down-regulated genes in the mutant plants to hormone and stress treatments. The expression ratios of the top 100 up-regulated genes in each plant (x-axes, from left to right) in response to the various hormone and stress treatments (y-axes) are displayed as heat maps.



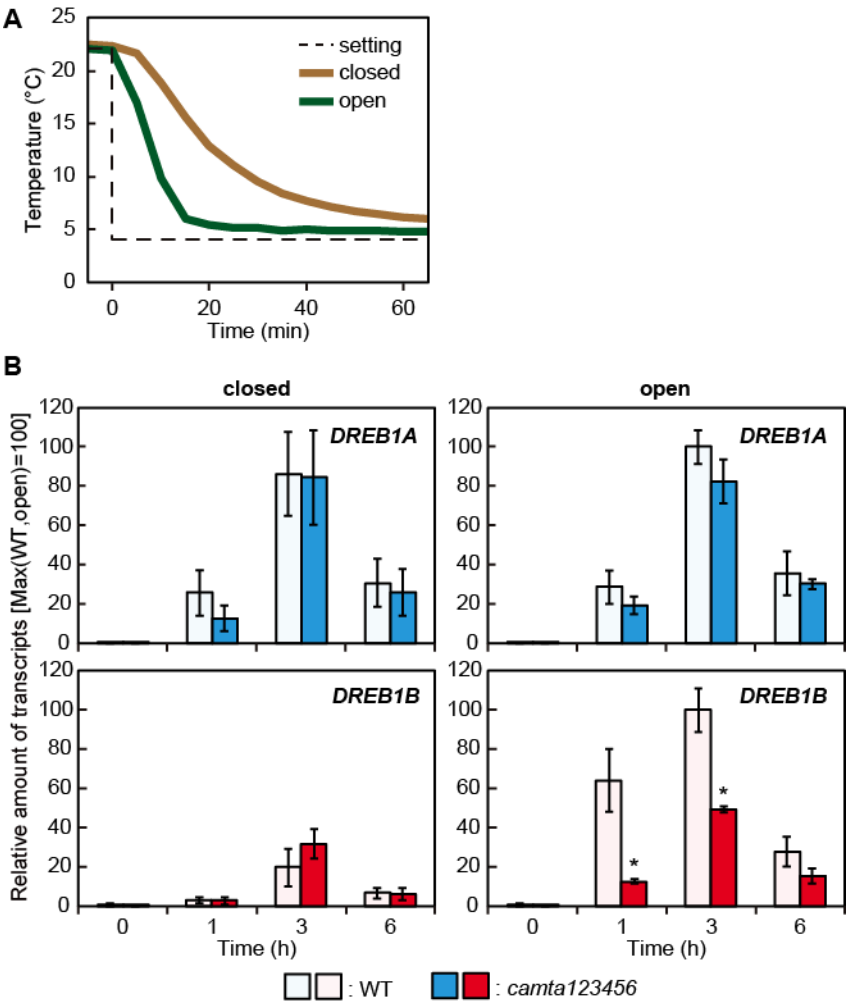
Supplemental Figure 7. Expression levels of the *CAMTA* family genes in Arabidopsis grown in soil pots or on agar plates.
(Supports Figure 4.)
The transcript level of each mRNA in 3-week-old Col-0 plants grown in soil pots or on agar plates at ZT2 under a 12-h light/12-h dark cycle at 22°C was measured by RT-qPCR. The values represent the average of three technical replicates, and the error bars indicate the SD. The expression level of each gene in the plants grown in soil pots was set to 1. The asterisks indicate significant differences (*$p < 0.05$ and **$p < 0.01$ according to Student's t-test) in the expression of each gene in the plants grown on agar plates, compared with that in the plants grown in soil pots in three biological replicates using the plants sampled at different times.



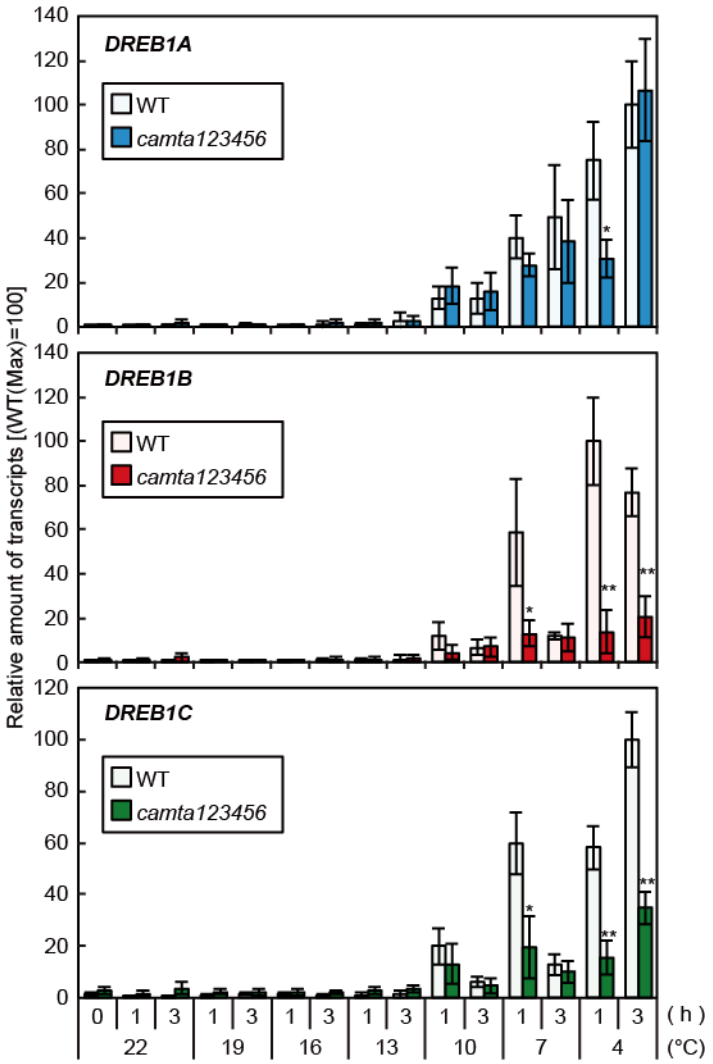
Supplemental Figure 8. Effects of cooling rates and growth conditions on cold-inducible expression of the *DREB1A* and *DREB1B* genes in various *camta* mutants.

(Supports Figure 5.)

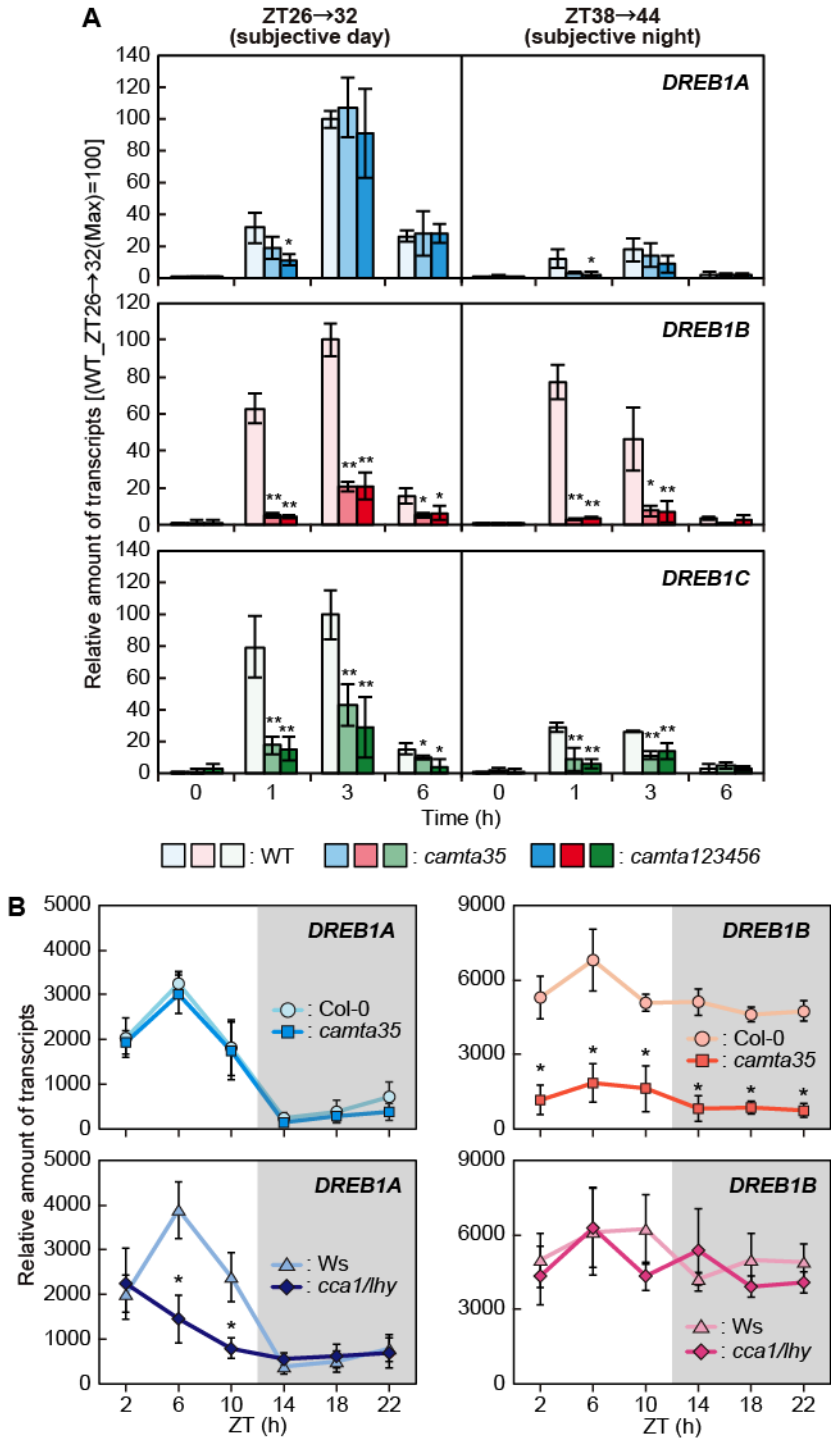
WT and *camta* mutant plants grown in soil pots were subjected to two different cold treatments (rapid and slow), as described in Figure 5A. The plants grown on agar plates were immediately cooled to 4°C. All treatments were carried out for 3 h after the temperature began to decrease. The transcript levels of each gene in the plants before (C) and after (L) the cold treatments were measured by RT-qPCR. The values represent the average of three technical replicates, and the error bars indicate the SD. The maximum expression level of each gene in the WT plants grown in soil pots under the rapid cooling treatment was set to 100. The letters above the bars indicate significant differences ($p < 0.01$ according to the Tukey–Kramer method) in the expression of each gene in three biological replicates using the plants sampled at different times.



Supplemental Figure 9. Cold-inducible expression of the *DREB1A* and *DREB1B* genes in plants grown on agar plates with or without lids.
(Supports Figure 5.)
(A) Temperatures around the plants grown on agar plates with or without lids.
(B) Expression levels of the *DREB1A* and *DREB1B* genes in response to cold stress in the plants grown on agar plates with or without lids. WT and *camta* sextuple mutant plants were grown on agar plates for three weeks. Then, they were immediately cooled to 4°C beginning at ZT2 with or without lids. The transcript level of each gene in the plants treated for the indicated duration (0, 1, 3, or 6 h) was measured by RT-qPCR. The values represent the average of three technical replicates, and the error bars indicate the SD. The maximum expression of each gene in the WT plants treated at 4°C without lids was set to 100. The asterisks indicate significant differences ($p < 0.01$ according to Student's t-test) in the expression of each gene compared with that in the WT plants at each time point in three biological replicates using the plants sampled at different times.



Supplemental Figure 10. Cold-inducible expression of the *DREB1* genes in plants grown in soil pots at various temperatures.
(Supports Figure 5.)
WT and *camta* sextuple mutant plants grown in soil pots were immediately treated at each temperature. The transcript level of each gene in the plants treated for the indicated duration (0, 1, or 3 h) beginning at ZT2 was measured by RT-qPCR. The values represent the average of three technical replicates, and the error bars indicate the SD. The maximum expression level of each gene in the WT plants was set to 100. The asterisks indicate significant differences (*$p < 0.05$ and **$p < 0.01$ according to Student's t-test) in the expression of each gene compared with that in the WT plants at each time point in three biological replicates using the plants sampled at different times.



Supplemental Figure 11. Effects of the diurnal cycle and circadian clock on cold-inducible expression of the *DREB1* genes.
(Supports Figure 6.)

Supplemental Figure 11. (continued)
(A) Expression of the *DREB1* genes in response to a rapid temperature decrease under circadian conditions. Three-week-old plants grown in soil pots under a 12-h light/12-h dark cycle at 22°C were transferred to free-running conditions under continuous light. Then, the plants were immediately cooled to 4°C beginning at Zeitgeber time 26 h (ZT26) or ZT38. The transcript level of each gene in the plants treated for the indicated duration (0, 1, 3, or 6 h) was measured by RT-qPCR. The values represent the average of three technical replicates, and the error bars indicate the SD. The maximum expression level of each gene in the WT plants treated at 4°C beginning at ZT26 was set to 100. The asterisks indicate significant differences (*$p < 0.05$ and **$p < 0.01$ according to Student's t-test) in expression at each time point compared with that in the WT in three biological replicates using the plants sampled at different times.
(B) Effects of the diurnal cycle on expression of the *DREB1A* and *DREB1B* genes in response to a rapid temperature decrease. Three-week-old plants grown in soil pots under a 12-h light/12-h dark cycle at 22°C were immediately cooled to 4°C for 1 h, every 4 h from ZT2 to ZT22. Several plants were treated and harvested at each time point. The values represent the average of three technical replicates, and the error bars indicate the SD. The transcript levels in the WT plants (Col-0 or Ws) before the cold treatment at ZT2 were set to 1. The asterisks indicate significant differences ($p < 0.01$ according to Student's t-test), in expression at each time point compared with that in the WT plants in three biological replicates using the plants sampled at different times.

Supplemental Table 1. The results of yeast one-hybrid screens (Supports Figure 1.)

Locus	Description	DNA binding domain	No.
At5g64220	CAMTA2	CG1	15
At1g22190	WIND2	AP2	1
At1g29860	WRKY71	WRKY	1
At2g02450	LOV1	NAM	1
At2g22120		zinc finger	1
At3g18650	AGL103	MADS-box	1
At3g19860	bHLH121	bHLH	1
At3g24140	FMA	bHLH	1
At5g10140	FLC	MADS-box	1
At5g22570	WRKY38	WRKY	1
At5g59820	ZAT12	zinc finger	1
			25

Supplemental Table 2. Enriched hexamers in the promoters of the genes downstream of of the CAMTA family genes.
(Supports Figure 3.)

	Sequence	List100	Random100		Fold	Z-score	*p*-value	*q*-value
			Mean100	SD				
UP	ACGACT	29	11.0	3.4	2.6	5.3	7.5E-08	3.1E-04
	AGTCGT	26	11.1	3.3	2.3	4.6	2.7E-06	4.1E-03
	AAAGTC	68	38.1	6.6	1.8	4.5	3.0E-06	4.1E-03
	GTCGTC	29	12.3	3.9	2.4	4.3	6.9E-06	7.0E-03
	TTCAAA	145	102.8	10.5	1.4	4.0	3.0E-05	2.4E-02
	A**TGAC**G	27	12.6	3.6	2.1	4.0	3.5E-05	2.4E-02
	AGACCG	18	7.3	2.8	2.5	3.8	6.4E-05	3.7E-02
	G**GTCA**A	42	23.0	5.0	1.8	3.8	7.2E-05	3.7E-02
	TCAAAA	160	117.7	11.3	1.4	3.7	9.1E-05	3.9E-02
	CGACTT	25	12.0	3.5	2.1	3.7	9.5E-05	3.9E-02
	CA**CGCG**	15	4.9	2.3	3.1	4.4	4.5E-06	1.9E-02
	A**CGCG**C	12	3.6	2.0	3.3	4.3	1.0E-05	2.1E-02
	TTACGC	18	7.2	2.7	2.5	4.1	2.5E-05	2.3E-02
DOWN	CCTATC	23	10.0	3.2	2.3	4.0	2.8E-05	2.3E-02
	CACGTG	40	19.8	5.0	2.0	4.0	2.8E-05	2.3E-02
	CAAATA	114	77.9	9.7	1.5	3.7	9.6E-05	6.0E-02
	AAAAAC	186	137.8	13.0	1.3	3.7	1.0E-04	6.0E-02
	CGCCAC	17	6.7	2.8	2.5	3.7	1.3E-04	6.5E-02
	TCCACG	24	11.4	3.5	2.1	3.6	1.4E-04	6.5E-02
	CACGAT	24	11.5	3.5	2.1	3.6	1.6E-04	6.6E-02

Red and blue letters indicated core sequence of W-box (5'-TGAC-3') and CGCG-box (5'-(A/C/G)CGCG(G/T/C)-3'), respectively.

Supplemental Table 3. Primers used in this study

Primer Name	Sequence
T-DNA insertion	
CAMTA1_LP	GAAAGTGACCATTCATCCACC
CAMTA1_RP	AAGGAAATGGACTTGATTGCC
CAMTA2_LP	TTTCAAAGAACCAACATTCATG
CAMTA2_RP	ACCTTGAAACCAATTCCTTGG
CAMTA3_LP	CTATGAGTGAACCAATTATCC
CAMTA3_RP	CAGTGAGCATGAAGTGCTGG
CAMTA4_LP	GATGCCCCTGAAGCTATAAGG
CAMTA4_RP	GTTGACCAATGTATGTTATC
CAMTA5_LP	GCTCTAGAGCTCTCCTCCGTAGC
CAMTA5_RP	CTACTAAGGAATTCCTCGGCG
CAMTA6_LP	CTGCAAGAGCATCCTTGAGAC
CAMTA6_RP	GCTTAACGATGGTCCACAAAG
LBb1.3	ATTTTGCCGATTTCGGAAC
plasmid construct	
CAMTA1pro_F_KpnI	GGGGTACCGTCGTAATTGGCCC
CAMTA1pro_R_XbaI	GCTCTAGAGATGGATAAACCCAAAAATC
CAMTA2pro_F_KpnI	GGGGTACCACGAATCAACGTTTC
CAMTA2pro_R_XbaI	GCTCTAGAGGAACATAAACCC
CAMTA3pro_F_KpnI	GGGGTACCGAGATGTATCTTTGTCATGAAC
CAMTA3pro_R_XbaI	GCTCTAGAAGATATTGCTTTCTC
CAMTA4pro_F_HindIII	GGGAAGCTTAATTTTGGGAAACATTGTG
CAMTA4pro_R_XbaI	GCTCTAGACTGAGAAGAAGAAAAAAACG
CAMTA5pro_F_HindIII	GGGAAGCTTAATAAATCTTATTTCTCC
CAMTA5pro_R_XbaI	GCTCTAGACATCTCCGGTTTCAGATAC
CAMTA6pro_F_KpnI	CGGGGTACCCTGTCCAAAAATGATTTCC
CAMTA6pro_R_XBaI	GCTCTAGACATCGTGATTTCACACTCG
CAMTA1_F_XbaI	GCTCTAGAATGGTGGATCGCAGATCTTTTG
CAMTA1_R_SmaI	TCCCCCGGGTCAAGGAGAAATAGACATC
CAMTA2_F_XbaI	GCTCTAGAATGGCGGATCGCGGATCTTTC
CAMTA2_R_SmaI	TCCCCCGGGTCATTCAAATGCAAGAGACATG
CAMTA2_R-3_SmaI	TCCCCCGGGTTCAAATGCAAGAGACATG
CAMTA3_F_XbaI	GCTCTAGAATGGCGGAAGCAAGACGATTC
CAMTA3_R_SmaI	TCCCCCGGGTTAACTGGTCCACAAAGATG
CAMTA3_CG-1_R_SmaI	TCCCCCGGGTCCATCCATGTCCCTC
CAMTA3_R-3_SmaI	TCCCCCGGGACTGGTCCACAAAGATGAGG
CAMTA4_F_SmaI	TCCCCCGGGATGGAATATGAAATTAGTAC
CAMTA4_R_SmaI	TCCCCCGGGTTGGTAACCTCGCACATGAG
CAMTA5_F_EcoRV	GGGGATATCATGGCCGGCGTTGATTCCGGC
CAMTA5_R_EcoRV	GGGGATATCTCAGCTCTCCTCCGTAGCC
CAMTA5_CG-1_R_SmaI	TCCCCCGGGTGAGTACGAGTTCCCAG
CAMTA5_R-3_EcoRV	GGGGATATCGCTCTCCTCCGTAGCC
CAMTA6_F_SmaI	TCCCCCGGGATGGACGGCGACGGTTTAGGC
CAMTA6_R_SmaI	TCCCCCGGGCTACAGATCTCTCGAGCC
EMSA	
DREB1Apro_WT_F_XbaI	GCTCTAGACGCTGTGTATAGTTTAC
DREB1Apro_WT_R_XbaI	GCTCTAGAACGGATATGTGGGGAG
DREB1Bpro_WT_F_XbaI	GCTCTAGACGCTATGTACTATAC
DREB1Bpro_WT_R_XbaI	GCTCTAGAACGGATATGTGGGGAG
DREB1Cpro_WT_F_XbaI	GCTCTAGAGCTGTTTCTTATCCAC
DREB1Cpro_WT_R_XbaI	GCTCTAGAGGATATTTGTGGGGTC

Primer Name	Sequence
quantitative RT-PCR	
CAMTA1_qRT_F	GCCTTCTAATACTGACTCAATGCTG
CAMTA1_qRT_R	TAACGCCAGATCAGTGTCATCC
CAMTA2_qRT_F	GCAACAAAATCCTGAACCTCAA
CAMTA2_qRT_R	GTTACTTCCTTTGACCCTGTTCC
CAMTA3_qRT_F	GCAACAGGAGGAAACCTTACTGG
CAMTA3_qRT_R	GTTACGGGGATTGTGCGAAG
CAMTA4_qRT_F	GCCGAATACCATTCTAGCAATCTC
CAMTA4_qRT_R	ATGCCTCCAACACATCTTTCC
CAMTA5_qRT_F	TCGTACTCAAGTTCGATCACTGAC
CAMTA5_qRT_R	AGACTGTTGCTCCGCACTTC
CAMTA6_qRT_F	CTGGATAAGGCTCGAGAGAACATC
CAMTA6_qRT_R	CGAGACGGAGATTGGACTAGAGA
DREB1A_qRT_F	CGCTGACTCGGCTTGGA
DREB1A_qRT_R	GCATCACACATCTCATCCTGAAAC
DREB1B_qRT_F(Ws&*cca1/lhy*)	GAGACGTGTGATACGACGACCA
DREB1B_qRT_R(Ws&*cca1/lhy*)	GCATCCCAAACATTGTCTCC
DREB1B_qRT-F	AGTCAACATGCGCCAAGGAT
DREB1B_qRT_R	ATGTCCAGGCCATGATTCG
DREB1C_qRT_F	TGACGTGTCCTTATGGAGCTA
DREB1C_qRT_R	CTGCACTCAAAAACATTTGCA
GolS3_qRT_F	ACAGGCCAAGAAGGAAATATGG
GolS3_qRT_R	GATGGAGCTTTGGCACATTG
COR15A_qRT_F	GAAAAAAACAGTGAAACCGCAGAT
COR15A_qRT_R	CCACATACGCCGCAGCTT
RD17_qRT_F	CAGTGTCGGAGAGTGTGGTG
RD17_qRT_R	ACAGCTGGTGAATCCTCTGC
COR15B_qRT_F	CCACAACGTAGGAGCAAAGCA
COR15B_qRT_R	TTCTTGCGCTGAGCAACGA
KIN2_qRT_F	CTGGCAAAGCTGAGGAGAAG
KIN2_qRT_R	ACTGCCGCATCCGATATACT
EDS1_qRT_F	CGAAGGGGACATAGATTGGA
EDS1_qRT_R	CGTAGCCTCTCTGAGCATATATG
ICS1_qRT_F	TATCTCCGGCAGCCGCCACT
ICS1_qRT_R	ACGCCGGAGGAAAACGACGG
PR1_qRT_F	TGATCCTCGTGGGAATTATGT
PR1_qRT_R	TGCATGATCACATCATTACTTCAT
XTH31_qRT_F	AGAGACCATACCCAAACACCAGA
XTH31_qRT_R	CCCGTCACAAGACCTCCAA
18S rRNA_qRT_F	AAACGGCTACCACATCCAAG
18S rRNA_qRT_R	CCTCCAATGGATCCTCGTTA
RT-PCR	
CAMTA1_F	ATGGTGGATCGCAGATCTTTTG
CAMTA1_R	GTGAAACATTACGCATGCCAG
CAMTA2_F	ATGGCGGATCGCGGATCTTTC
CAMTA2_R	GATTGCATCTGCAAGTCTTC
CAMTA3_F	= CAMTA3_RP
CAMTA3_R	TTAACTGGTCCACAAAGATG
CAMTA4_F	GTTGACCAATCTACATTTTTGTTG
CAMTA4_R	TTGGTAACCTCGCACATGAG
CAMTA5_F	= CAMTA5_RP
CAMTA5_R	TCAGCTCTCCTCCGTAGCC
CAMTA6_F	= CAMTA6_RP
CAMTA6_R	CTACAGATCTCTCGAGCC
Act2_F	GGAAGGATCTGTACGGTAAC
Act2_R	TGTGAACCATTCCTGGAC

An underline indicates a restriction site.