Table S1. Gene-specific primer pairs used in this study

Gene	Primers (5' to 3')
For protein expression construct	
ALKBH6	Forward: GGATCCATGGAGTTAGAGAGATTCAGG Reverse: GTCGACGAATCTGAAGAGATTTTTACG
For genomic DNA PCR	
ALKBH6	Forward: TCATTCTCTGTCTTGTTGATGCC Reverse: CCAAAGCTTCAGCTTCATTAACG Forward: TTCCTCCACGGCATTAGCGATAG Reverse: ACGGACTTTGGGAACTAGACGGC
For RT-PCR	
ALKBH6	Forward: TCAAGATGGACCCGCTTACTTTCC Reverse: ACGGACTTTGGGAACTAGACGGC
For quantitative real-time RT-PCR	
ALKBH6	Forward: TGTCTCTTGGCTCACCTGTTGT Reverse: ACGGACTTTGGGAACTAGACGGC
RD29A	Forward: CGGAGGAAATTATTCCACCAGGGA Reverse: ACAGAATGAGCCGGTGCATCGTG
RD29B	Forward: TCCTGTCGTGTCTTCTGACCACAC Reverse: TCGCTTCCCAGTCCGATGTTTCC
CBF2	Forward: GGTCTTGACATGGAGGAGGAGACC Reverse: CAACATCGCCTCTTCATCCATA
SUS	Forward: TAAGCTCTTCCATGACAAGGAGA Reverse: AAGGTGTGTTGCAGAGTGTTGA
HSP70	Forward: GGGCACGAACAAAGGACAAC Reverse: GCCCAGCTCCTTGGTACATT
ABA1	Forward: GAACGTACTATAAAGGGAGAAGG Reverse: CTGAGACGAAGGGATCACAAT
ABA2	Forward: CTAAACTCGCTTTGGCTCATT Reverse: CGCTACATCATCAACCGTCAG
ABA3	Forward: TTTGCTGGGATGACAATGAT Reverse: CCTTCCACTGACGACGGTTC
NCED3	Forward: TAAAAACCGTTGGTCGGTTC Reverse: CGACGTCCGGTGATTTAGTT
ABI1	Forward: AGAGTGTGCCTTTGTATGGTTTA Reverse: CATCCTCTCTCTCTACAATAGTTCGCT
ABI5	Forward: CAATAAGAGAGGGATAGCGAACGAG Reverse: CGTCCATTGCTGTCTCCTCCA
SnRK2D	Forward: TTGTTATGGAATATGCTGCTGGTGG Reverse: TCAGATCCCGATGGCATATTTGC
SnRK2E	Forward: AACTCCTGCTTAATCGCTCCTAGAG Reverse: TTTCCTGAAATTCTTTGGTCCTCG
SnRK2I	Forward: GCAATGCAGGACGGTTTAGTGAAG Reverse: TTAATCGAGGAGCAGGACTTCCATC
SnRK2-6	Forward: TTTGTTGCTGACCCTGCAAAGAGG Reverse: TCTTCTATGCTTTGGCCCGGTTGA
ABF3	Forward: GGTTGATGATGTCTGGAAGGAGC Reverse: CCCTAACCACACCAGCCCTGA
PYR1	Forward: GCGAACACATCAACGGAAAG Reverse: CCAGATCCGATTCTCTTTCTCG

Supplementary Figure S1. Domain structures of the *ALKB* domain-containing proteins in *Arabidopsis.* Full-length amino acid sequences of the thirteen ALKB domain-containing proteins were downloaded from the *Arabidopsis* genome sequence database (http://arabidopsis.org) and analyzed using a SMART program (http://smart.embl.de). Blue bars represent the conserved 2-oxoglutarate (2OG) and Fe(II)-dependent oxygenase (Oxy) domains.

Supplementary Figure S2. The expression patterns of the stress-responsive marker genes in *Arabidopsis*. Two-weeks-old *Arabidopsis* Col-0 seedlings were subjected to drought, heat (42^{o}C), cold (10^{o}C), salt (300 mM NaCl) stress, or ABA treatment (100 μM ABA) for the indicated times, and the expression levels of each gene were determined via quantitative real-time RT-PCR. Values are the mean ± SE obtained from three independent experiments.

Supplementary Figure S3. Confirmation of the T-DNA insertion *alkbh6* mutants. (A) Schematic representation of the T-DNA insertion site. Exons and introns are represented by black boxes and lines, respectively. The position of T-DNA insertion in the mutant is indicated by a black triangle. (B) Genomic DNA PCR analysis to confirm the homozygous line of the mutant. (C) RT-PCR analysis to show the down regulation of *ALKBH6* expression in the mutants. Tubulin was used as a loading control. (D) Real-time RT-PCR analysis to confirm the reduced level of *ALKBH6* in the mutants. Values are the mean ± SE obtained from three independent experiments.

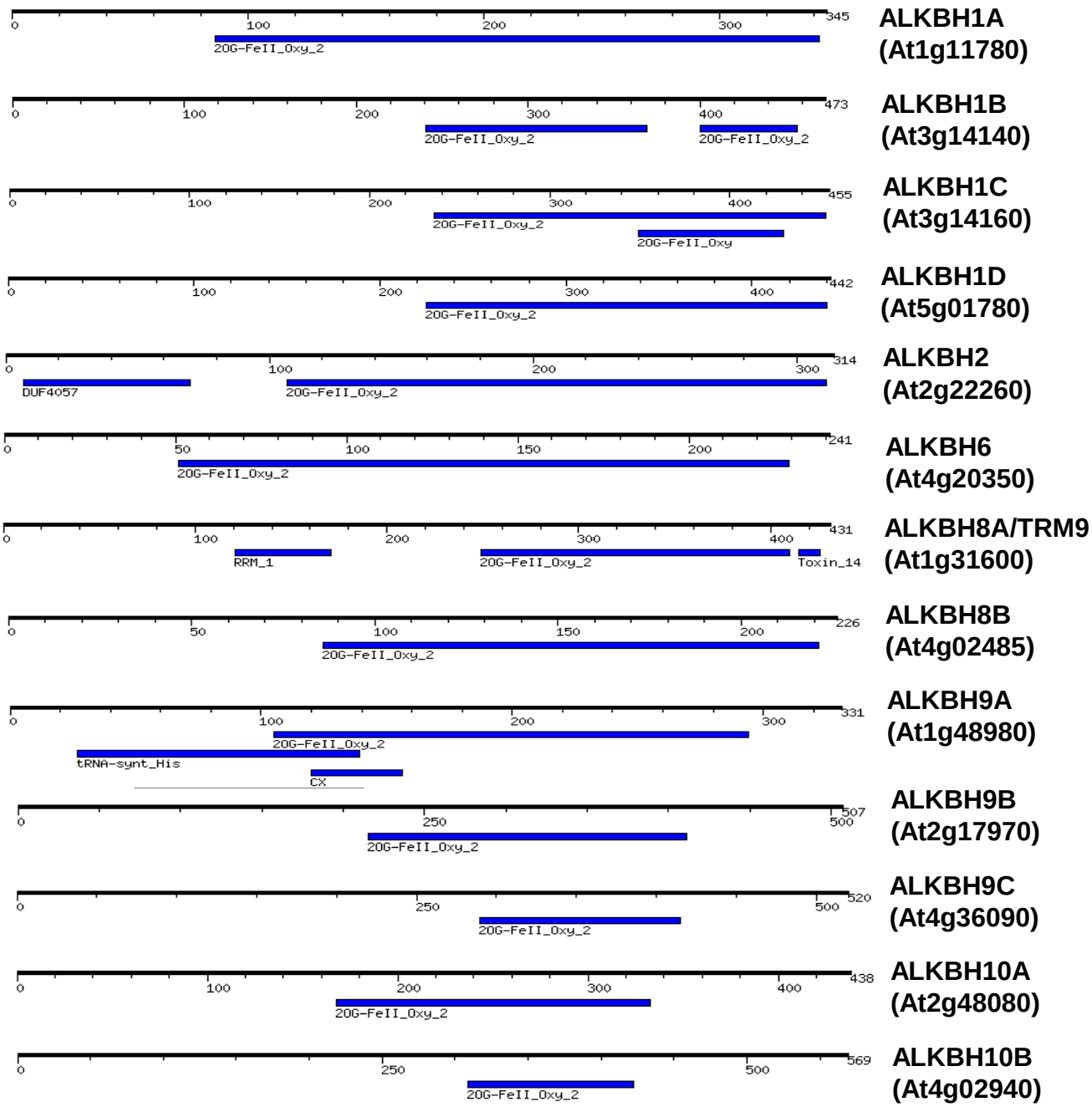
Supplementary Figure S4. Seed germination of the *alkbh6* mutants under normal and abiotic stress conditions. Germination percentages of the wild type and *alkbh6* mutants were scored on the MS medium supplemented with different concentrations of mannitol, NaCl, or ABA, and on MS medium at cold temperature (10℃). The mean values and standard errors (bar) were obtained from three independent experiments.

Supplementary Figure S5. Phenotypes of the *alkbh6* mutants under normal growth conditions. Growth of the wild type and *alkbh6* mutants (A) on MS and (B, C) in soil under normal growth conditions. DAG, days after germination.

Supplemental Figure S6. Drought sensitivity of the *alkbh6* mutants. Twenty-days-old seedlings grown in soil were subjected to drought stress for 13 days, and survival rates were measured 3 days after recovery.

Supplementary Figure S7. Levels of m^{6}A/A and m^{5}C in the *alkbh6* mutants. Total RNA in 18-days-old wild type (WT) and *alkbh6* mutant (6-1 and 6-2) seedlings was extracted, and the levels of m^{6}A/A (%) and m^{5}C (%) were determined using an EpiQuik m^{6}A RNA Methylation Quantification Kit and a MethylFlash 5-mC RNA Methylation ELISA Easy Kit, respectively. Values are the mean ± SE obtained from three independent experiments. NC, negative control; PC, positive control.

Supplementary Figure S8. Purification of the recombinant ALKBH6 protein. The His-tag-ALKBH6 fusion protein was expressed in *E. coli* and purified using a Ni-NTA His•Bind resin. M, molecular size marker; U, un-induced; I, induced; P, purified.



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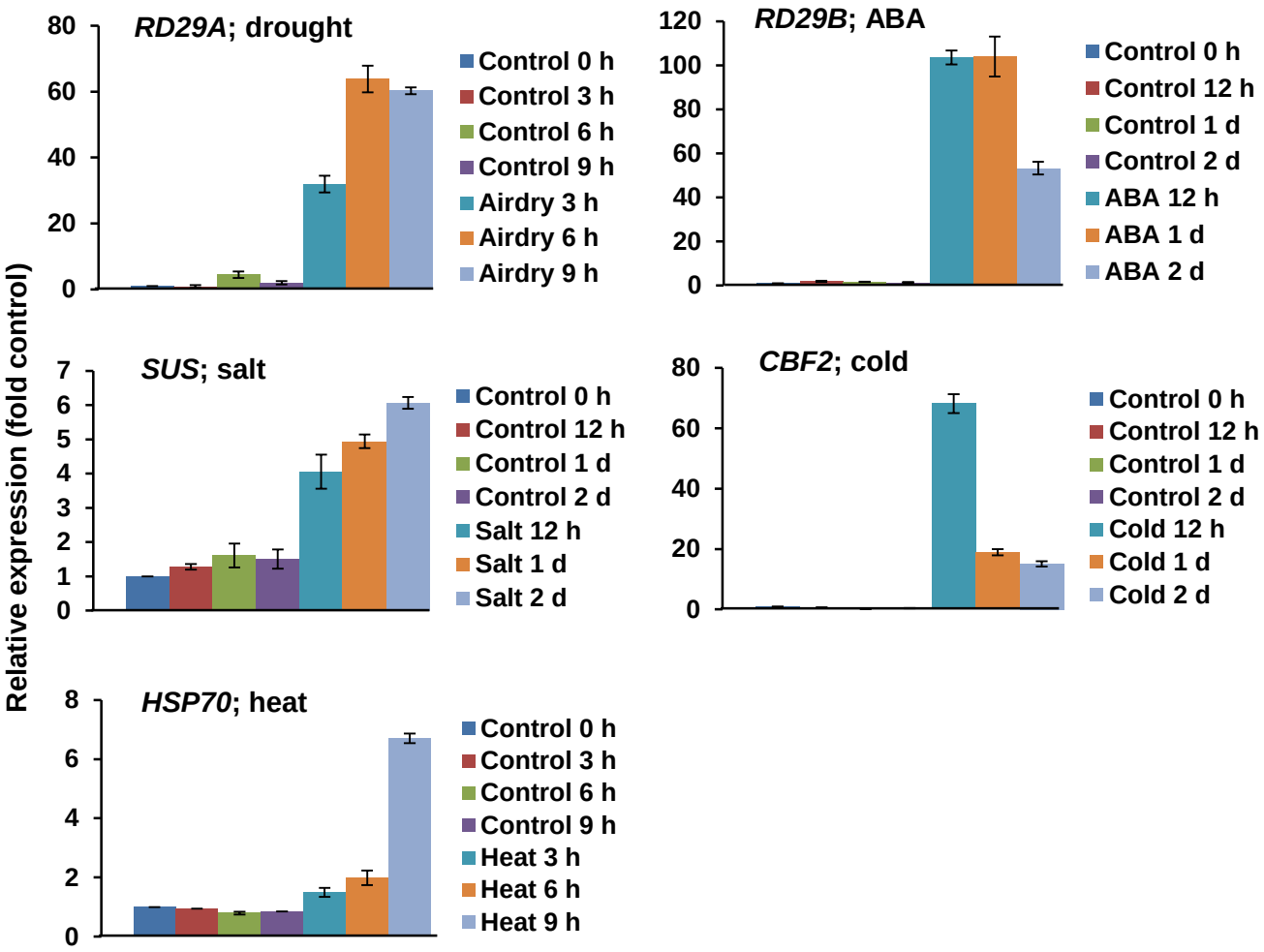

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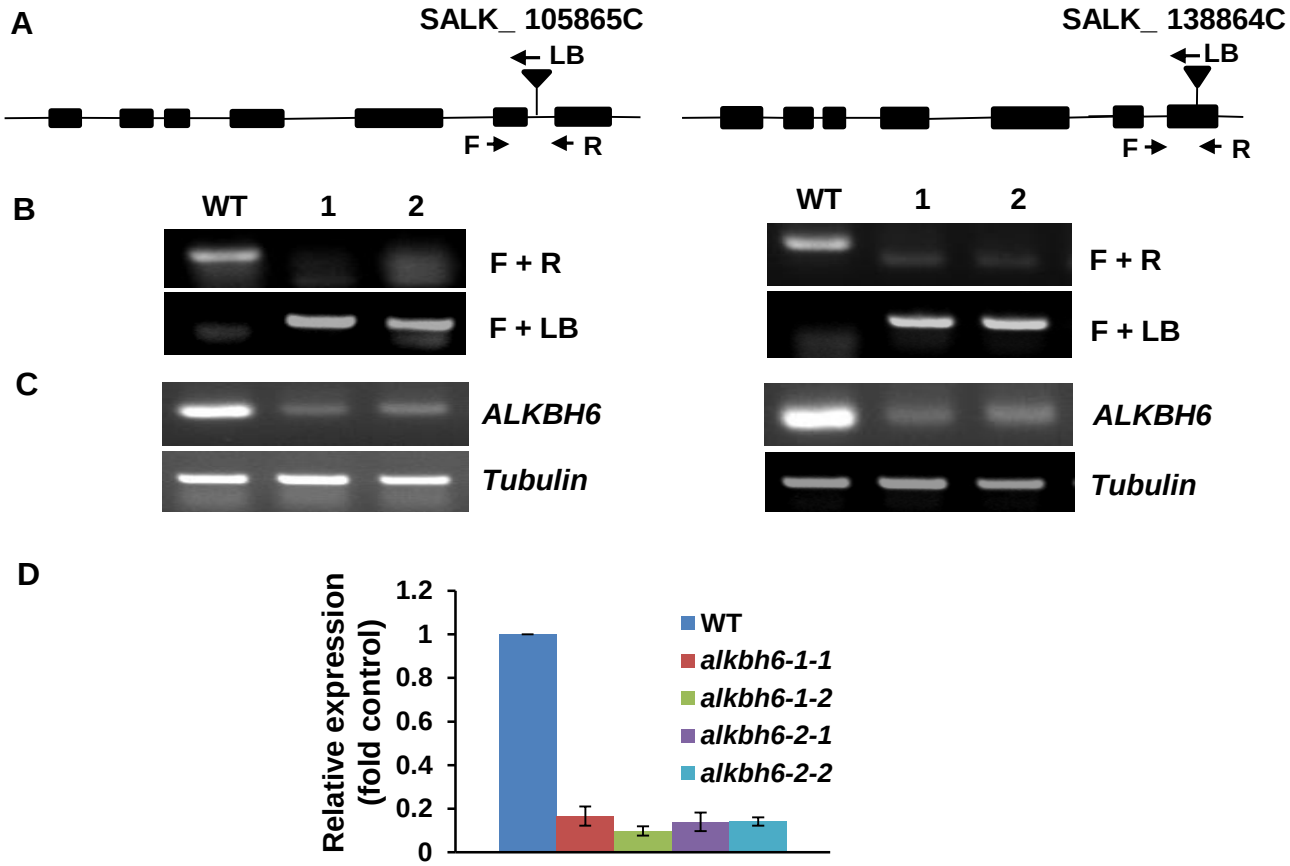

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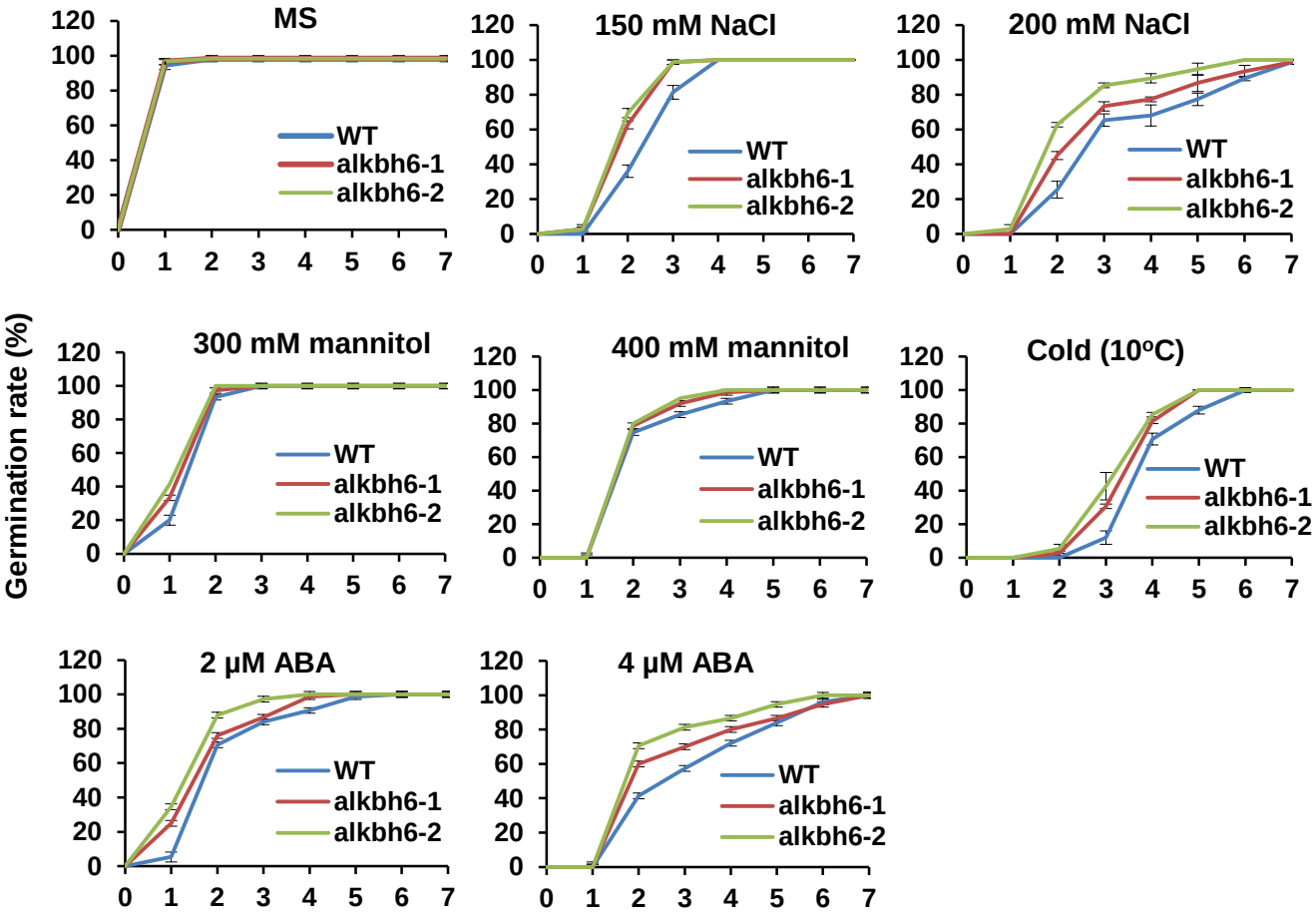

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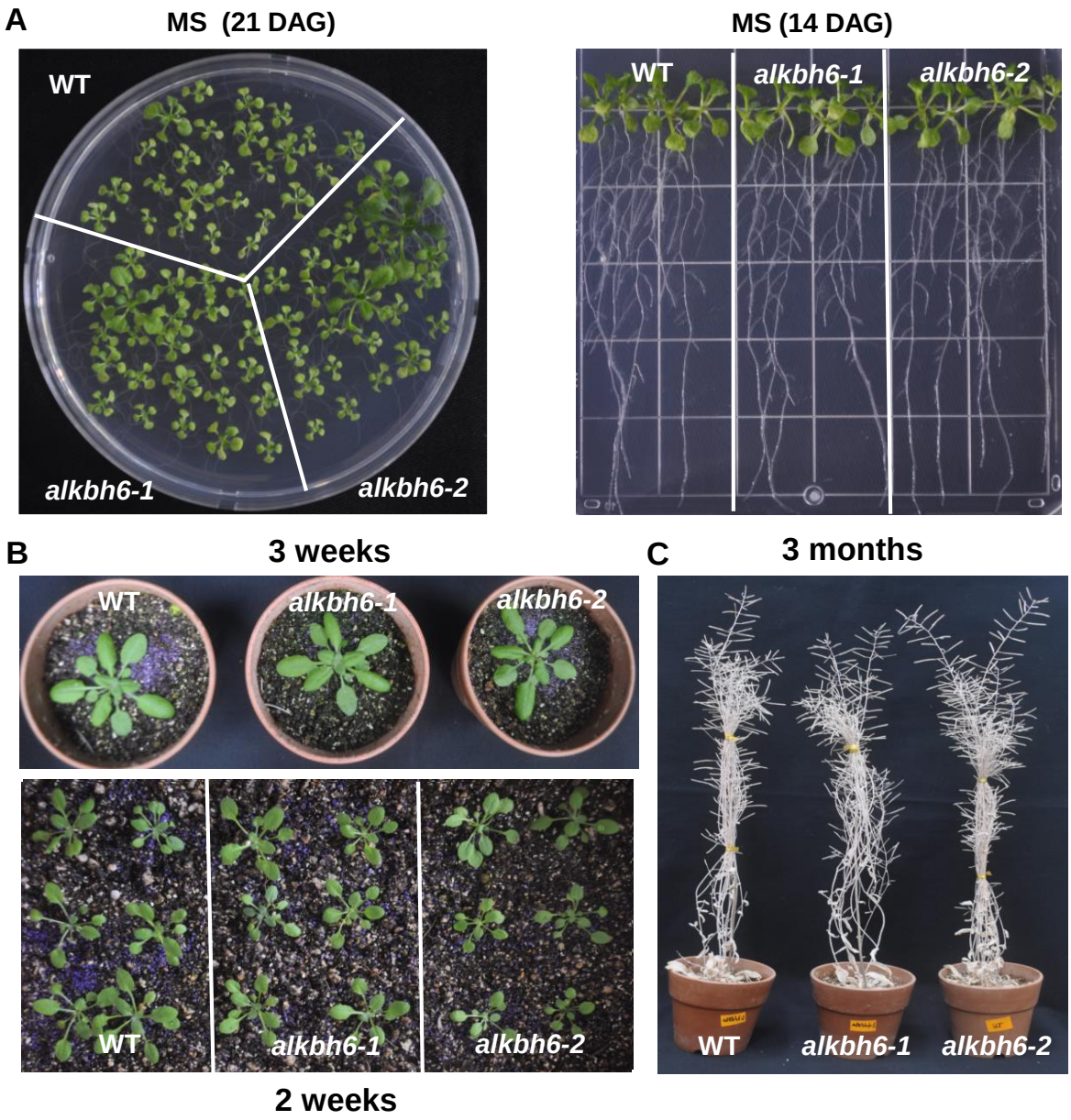

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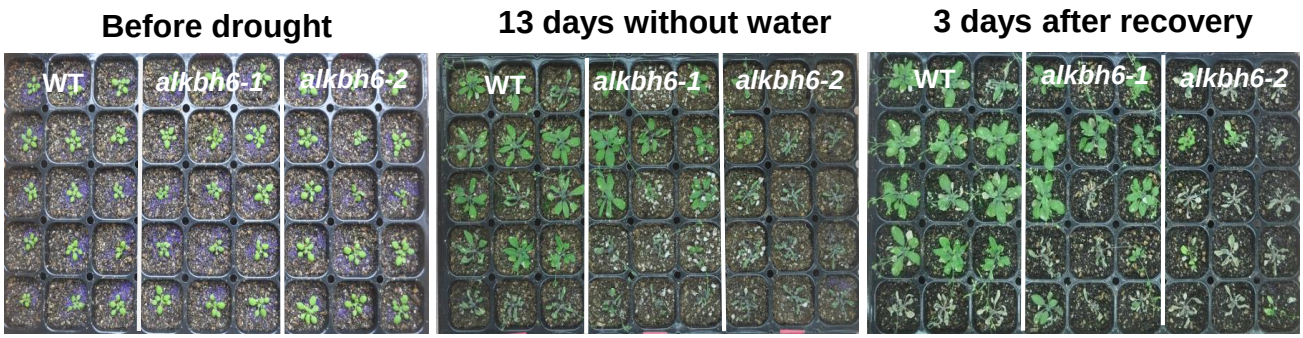

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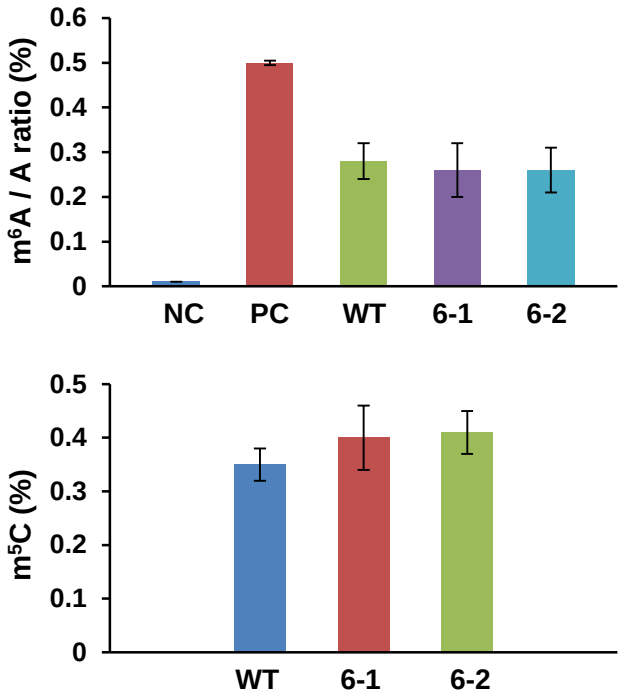

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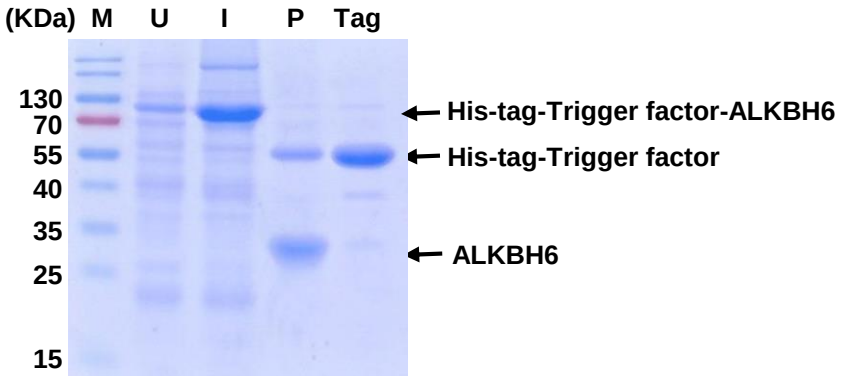

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