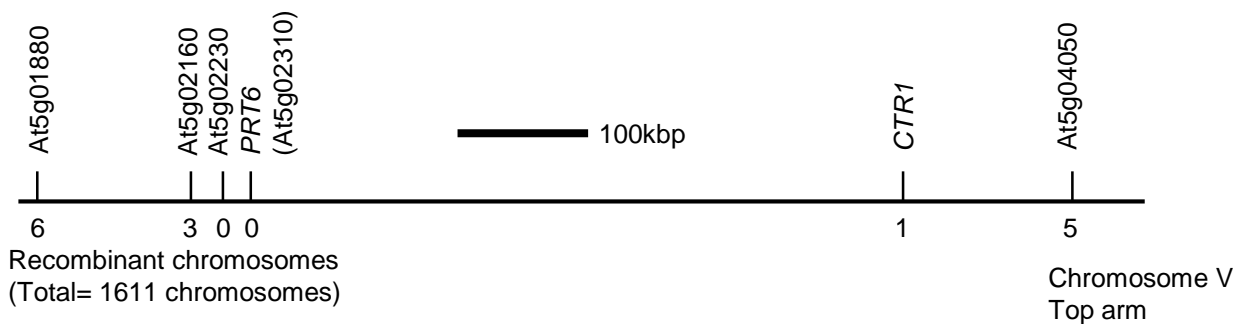


Supplementary Figure 1:

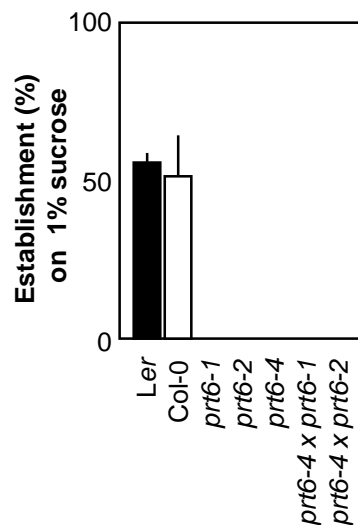
A.



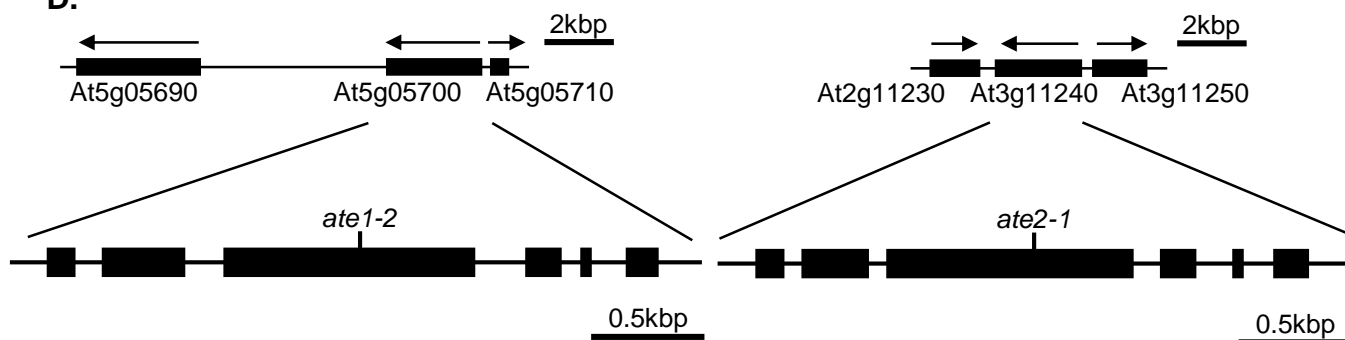
B.

Genotype:	ABA (μM)		
	0 (SE)	0.5 (SE)	5 (SE)
<i>prt6-1</i> (Col-0)	94 (4)	4 (2)	0 (0)
<i>prt6-2</i> (Col-0)	100 (0)	2 (1)	0 (0)
Col-0	99 (1)	95 (1)	2 (2)
<i>prt6-4</i> (Ler)	98 (2)	4 (1)	0 (0)
Ler	95 (2)	80 (10)	1 (1)
<i>prt6-4</i> x <i>prt6-1</i>	93 (0)	0 (0)	0 (0)
<i>prt6-4</i> x <i>prt6-2</i>	100 (0)	0 (0)	0 (0)

C.



D.



Supplementary Figure 1: Positional cloning of *prt6-4*.

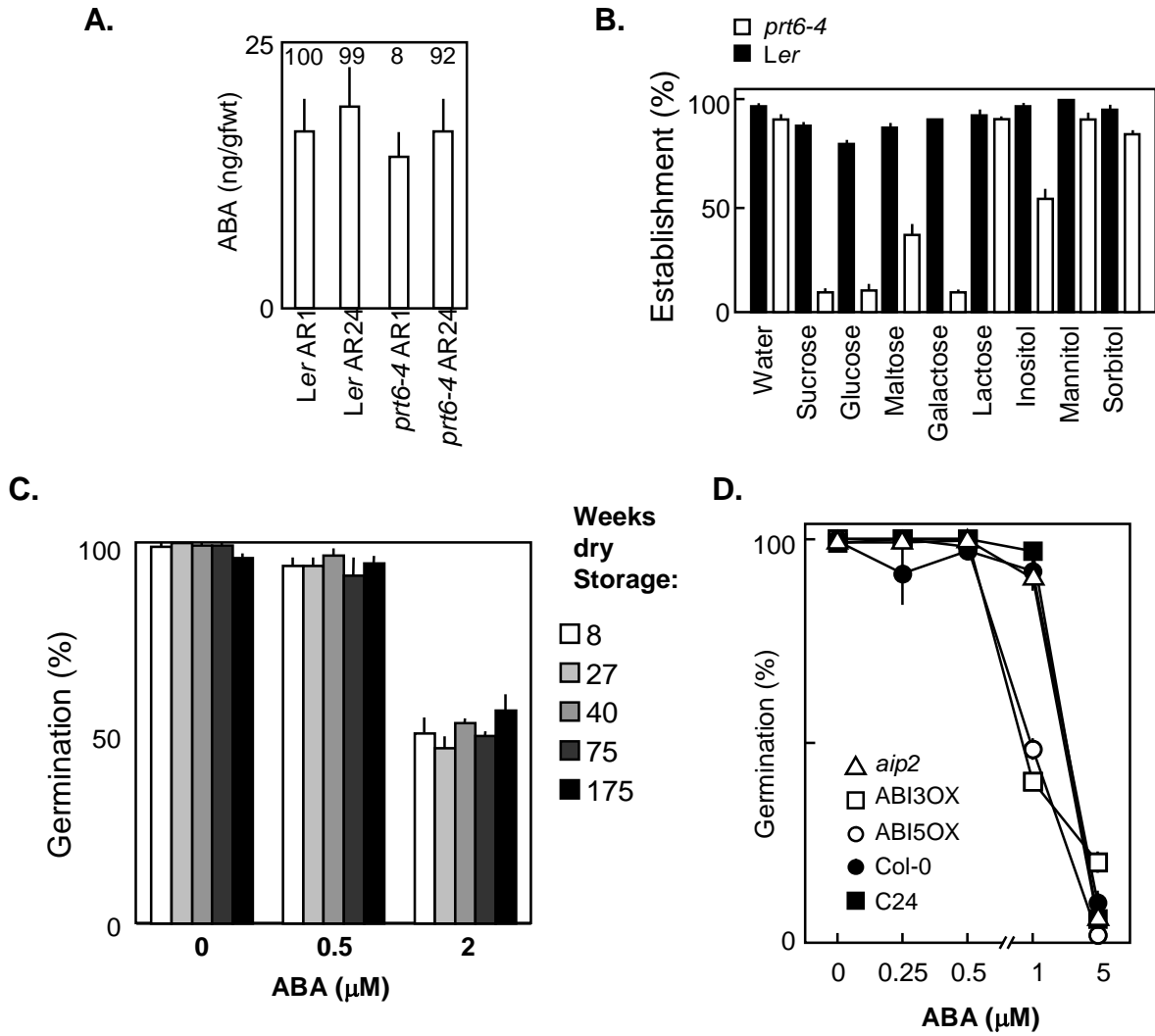
A. Chromosomal positions of molecular markers used to map the position of the mutation represented by *prt6-4* (originally denoted in our genetic screen as fast neutron line 10; *fn10*). An F2 population was developed between *Ler* and *Col-0*, and used to map the position of the mutation. Numbers of recombinant chromosomes in the mapping population are indicated. A diagrammatic representation of the *PRT6* gene (At5g02310) shows the position of the *prt6-4* (*fn10*) mutation, that is an insertion 497178 kbp from the top of chromosome V into the *PRT6* gene. The positions of other previously identified *prt6* alleles are also shown (all T-DNA insertions).

B,C Complementation analysis demonstrates that the *prt6-4* (*fn10*) mutation is in the same genetic complementation group as *prt6-1* and *prt6-2*.

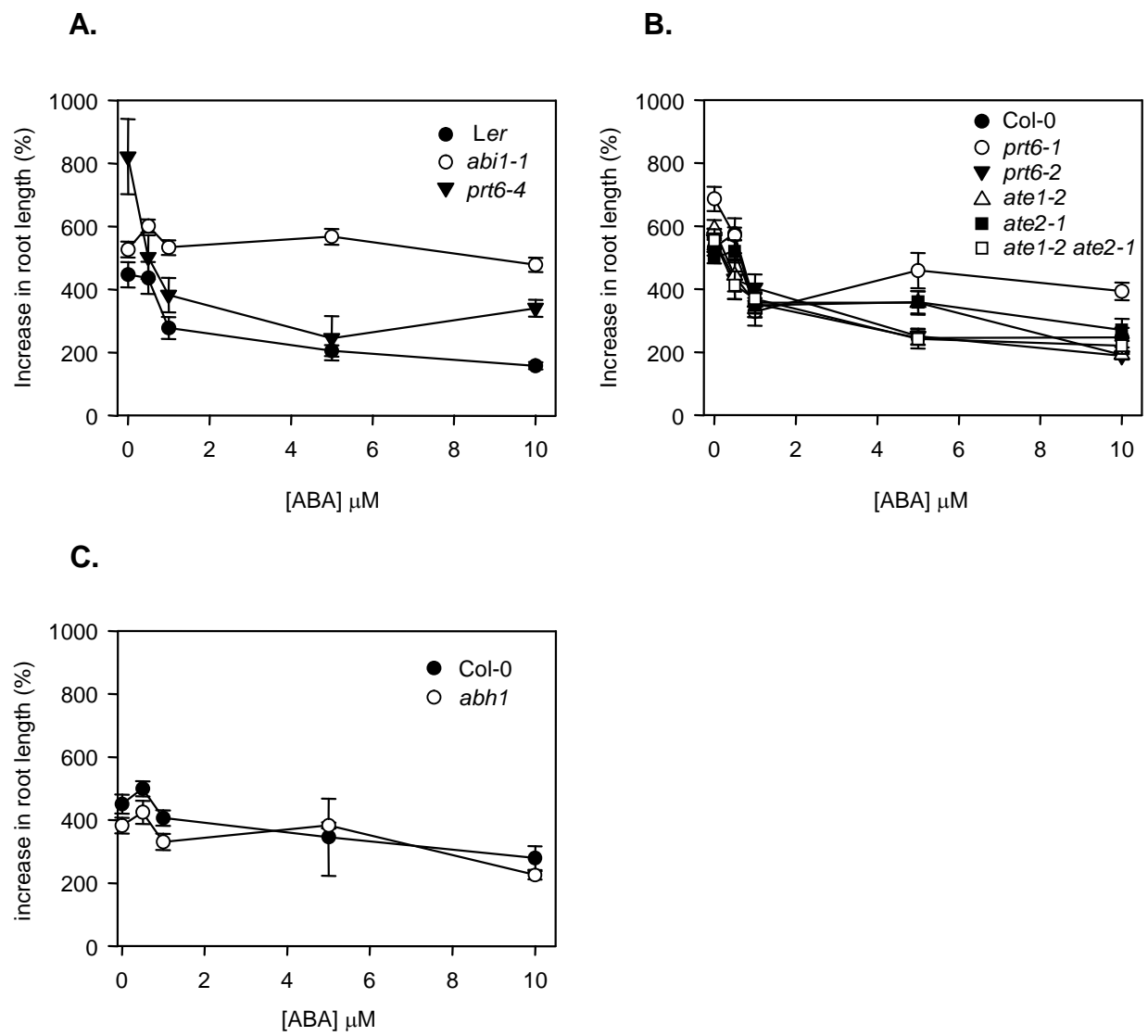
B. Reduced ABA sensitivity cannot be restored in crosses between *prt6* alleles. Therefore increased ABA sensitivity of all mutants is due to disruption of the the same gene (*PRT6*). Germination potential of moist chilled seeds on 1/2 MS supplemented with ABA is indicated, following 7 days imbibition. Data points represent mean values in percent +/- standard error of the mean (SE).

C. Reduced sugar sensitivity cannot be restored in crosses between *prt6* alleles. Therefore increased sugar sensitivity of all mutants is due to disruption of the the same gene (*PRT6*). Establishment of moist chilled seeds on water agarose supplemented with 1% (w/v) sucrose 7 days imbibition following 2 days moist chilling. Data points represent mean values in percent +/- standard error of the mean (SE).

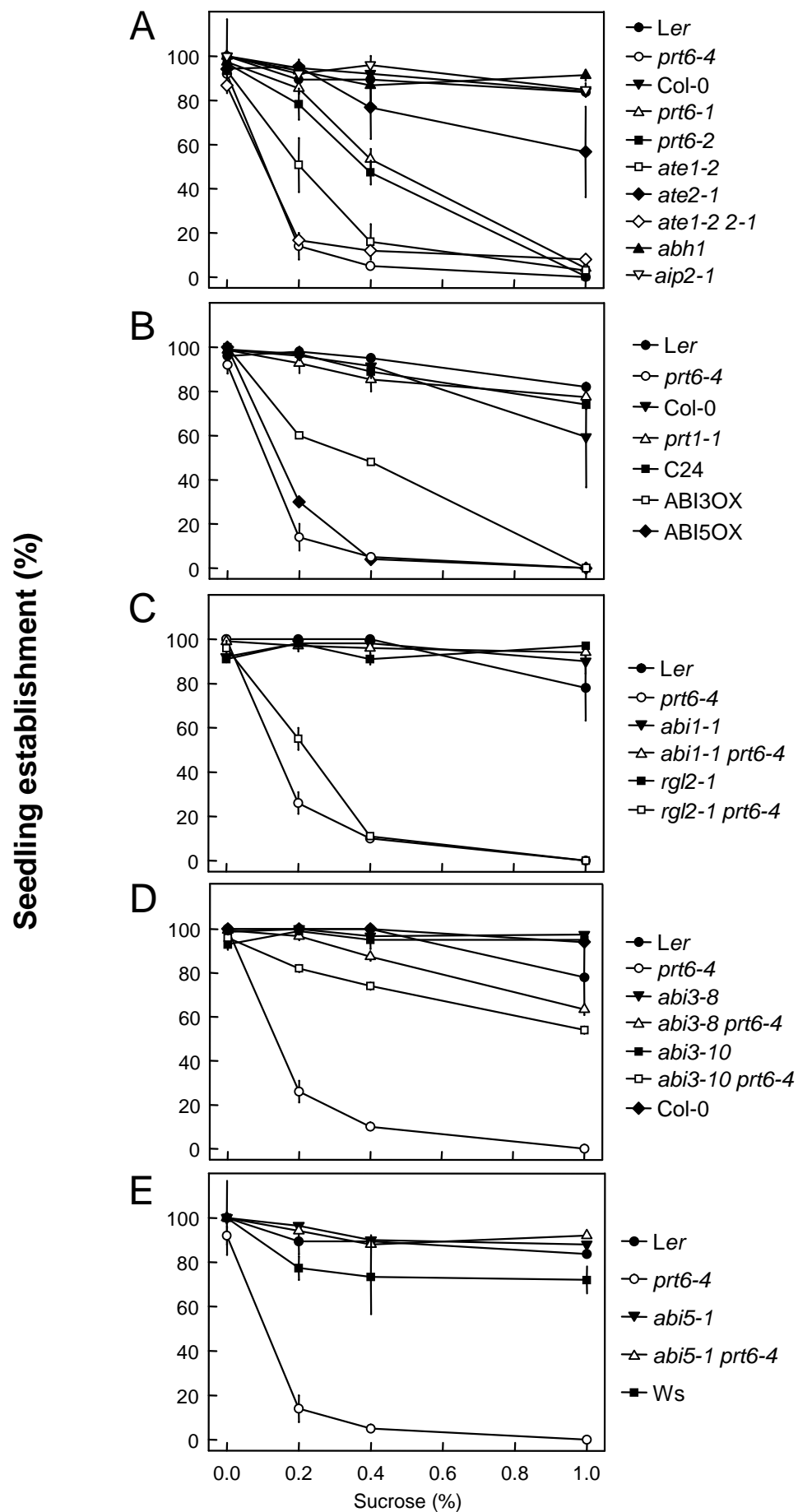
D. Positions of TDNA insertions in genes *ATE1* and *ATE2*. *ate1-2* corresponds to SALK_023492, and *ate2-1* to SALK_040788.



Supplementary Figure 2: Physiological characteristics of *prt6-4* mutant seeds.



Supplementary Figure 3. Mutants in the N-end rule pathway are not hypersensitive to ABA for root elongation.



Supplementary Figure 4: Effect of exogenous sucrose on seedling establishment of single and double mutants

Supplementary Figure 4: Effect of exogenous sucrose on seedling establishment of single and double mutants

Seeds were plated on water agarose media containing sucrose, as indicated. Establishment was scored as the presence/absence of green cotyledons, after 7 days. Data represent means \pm SE of the mean.

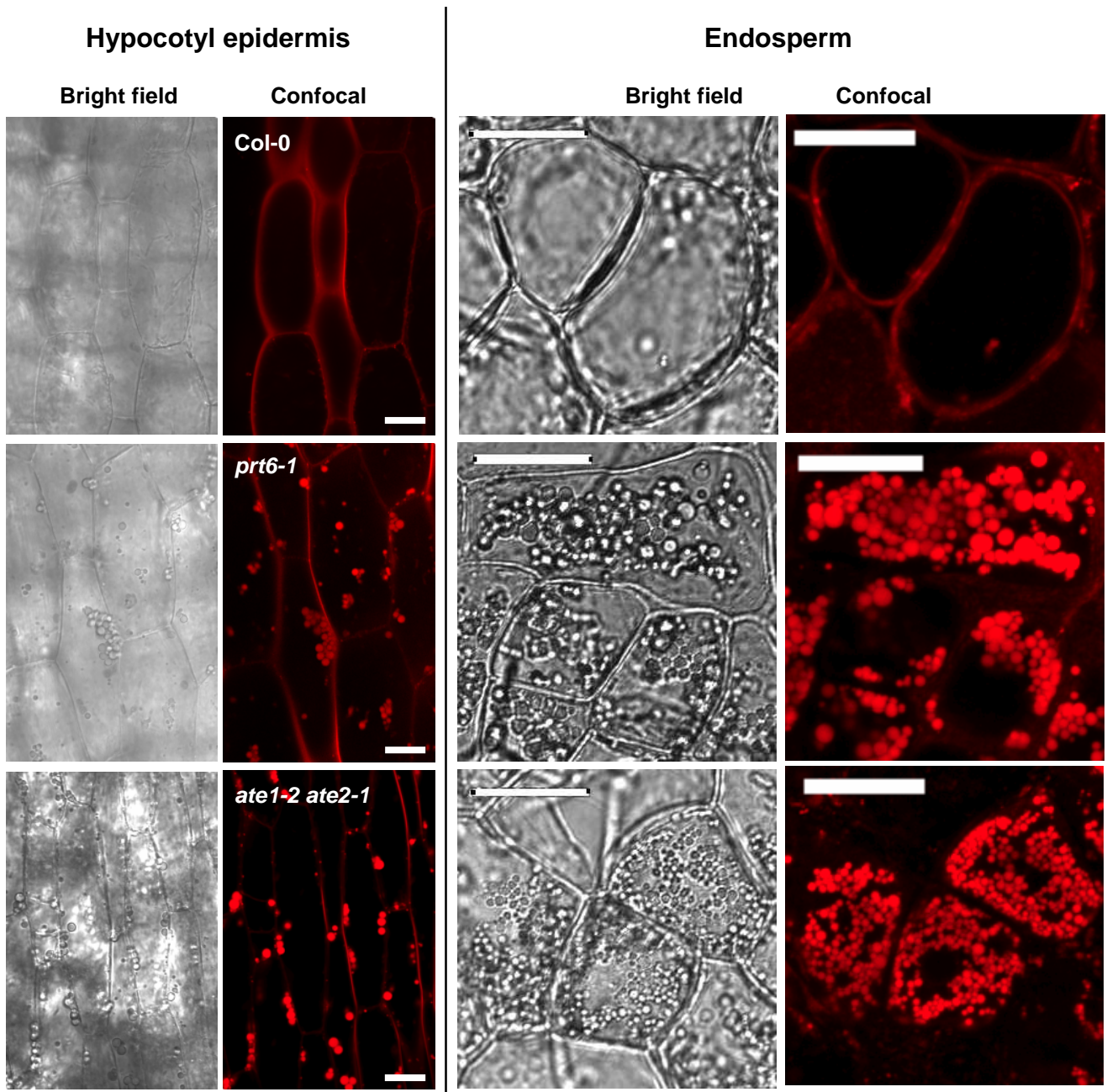
A. N-end rule pathway mutants (*prt6-4*, Ler background; *prt6-1*, *prt6-2*, *ate1-2*, *ate2-1*; Col-0 background) and the ABA hypersensitive mutants, *abh1* and *aip2-1* (Col-0). *abh1* and *aip2-1* are insensitive to exogenous sucrose, *ate2-1* shows modest sensitivity, whereas establishment of *prt6-1*, *prt6-2*, *prt6-4*, *ate1-2* and the *ate1-2 ate2-1* double mutant is sensitive to applied sugar.

B. Seedling establishment of the N-end rule pathway mutant, *prt1-1* is insensitive to exogenous sucrose, as are transgenics which express ABI3 and ABI5 under the control of the 35SCaMV promoter (*ABI3OX*, Col-0; *ABI5OX*, C24 background, respectively). Ler and *prt6-4* are shown for comparison.

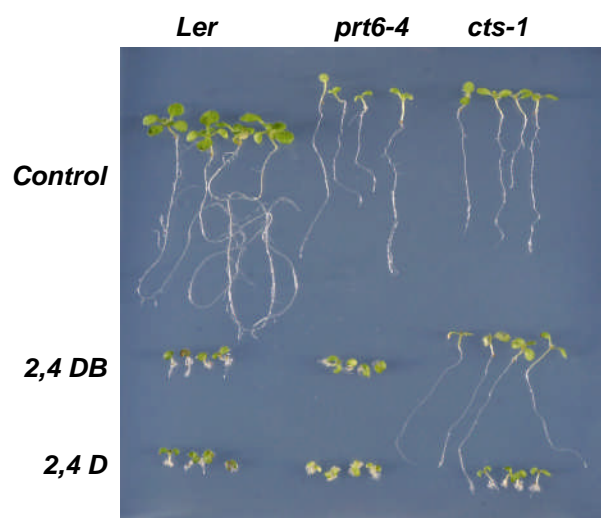
C. Genetic interactions of *prt6-4* with components of ABA synthesis and signalling (*abi1-1*, *aba1-1*; Ler background) and GA signalling (*rgl2*; Ler background).

D. Genetic interactions of *prt6-4* with ABI3. *abi3-8* and *abi3-10* alleles are in the Col-0 background.

E. Genetic interactions of *prt6-4* with ABI5. *abi5-1* is in the Ws background. ABI5 is epistatic to PRT6 in this assay.

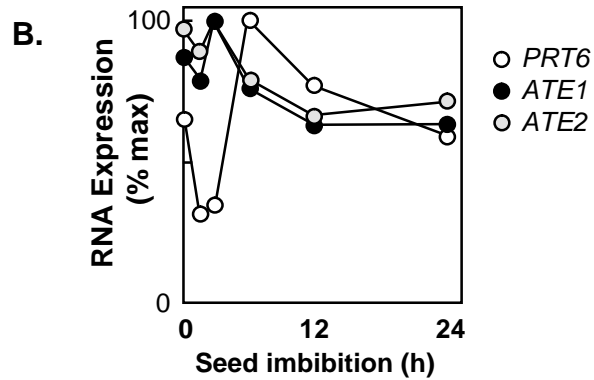
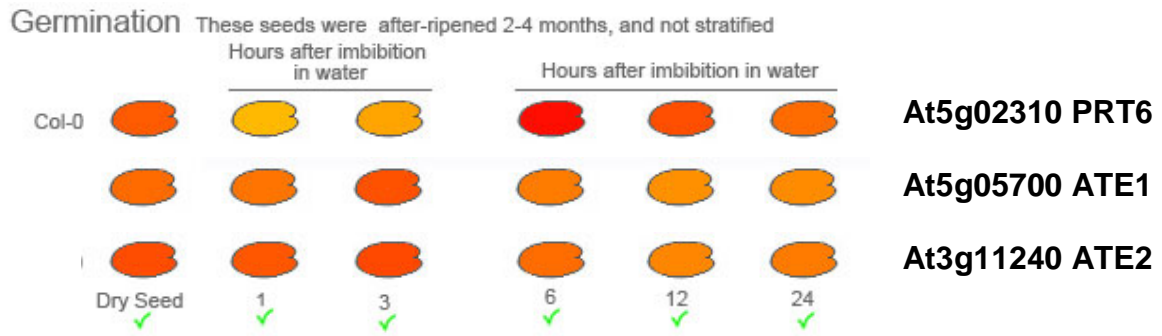


Supplementary Figure 5:
Bright field and confocal microscopy reveal oil body retention in hypocotyl epidermis and endosperm of N-end rule pathway mutants.

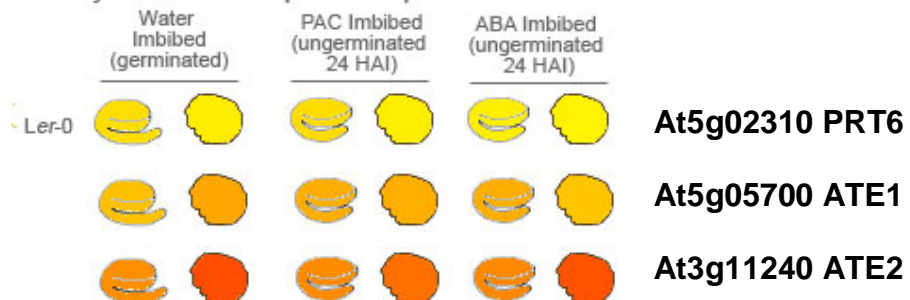


Supplementary Figure 6. The *prt6-4* mutant is sensitive to 2,4-DB.

A.



C. Embryo and Endosperm Separated



Supplementary Figure 7:

Relative expression of RNA for *PRT6*, *ATE1* and *ATE2* during seed germination.