Supporting Information

Guo et al. 10.1073/pnas.0812346106

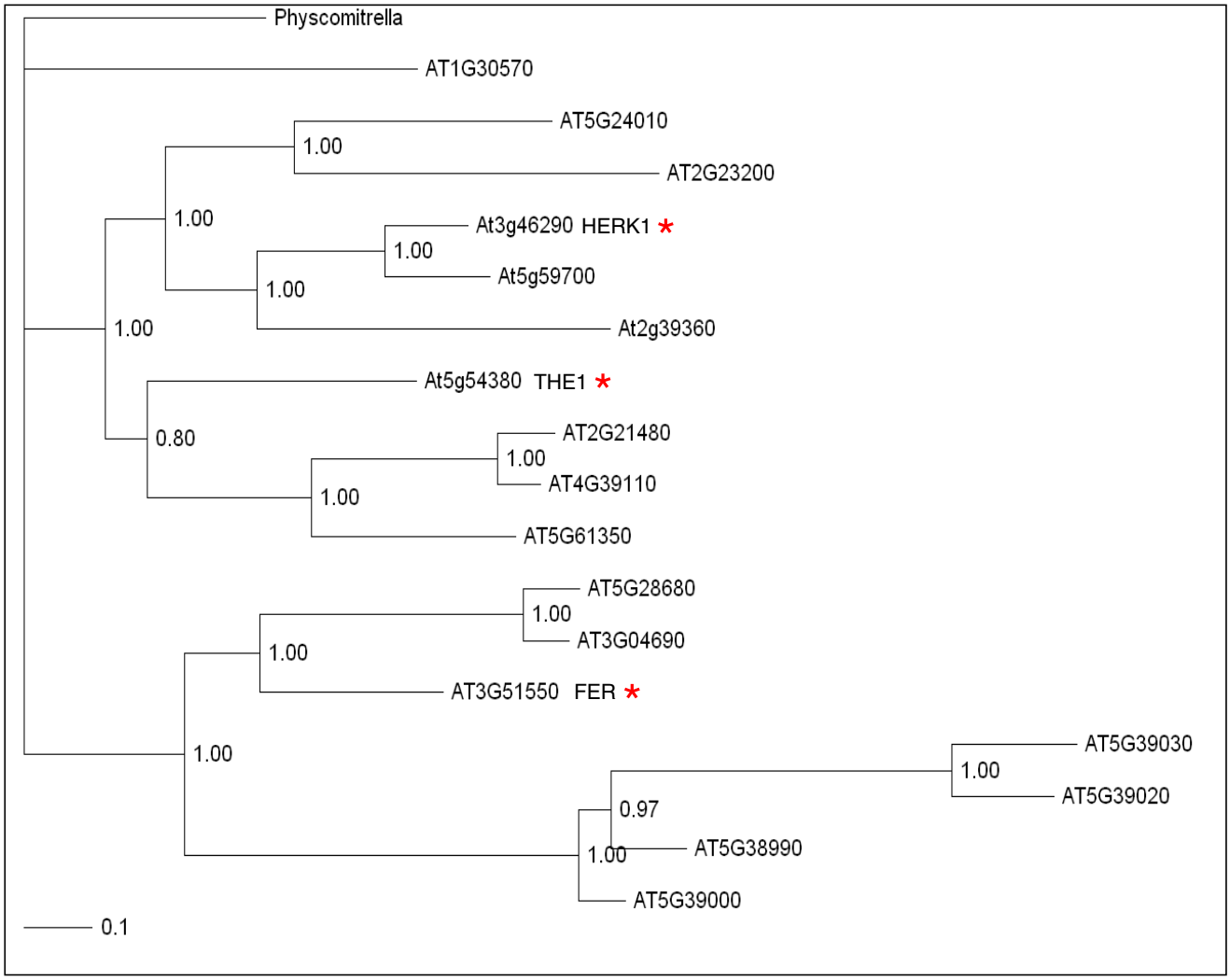

Fig. S1. Arabidopsis HERK1, THE1 and FER are related RLKs in the CrRLK family. We obtained all protein sequences from NCBI, which were then aligned by ClustalX 2.0 (http://bips.u-strasbg.fr/fr/Documentation/ClustalX). Mrbayes program (http://mrbayes.csit.fsu.edu/index.php) was used to reconstruct the evolutionary tree of 17 CrRLK proteins and the tree was viewed by TreeViewX (http://darwin.zoology.gla.ac.uk/~rpage/treeviewx). A CrRLK homolog in *Physcomitrella patens* (XP_001760700) was used as the outgroup during the analysis. After 500,000 iterations in Mrbayes, the split frequency reached as low as 0.005, which indicates high confidence that convergence occurred. The numbers on the tree indicated the posterior probability of each clade (bigger posterior probability indicates higher confidence).

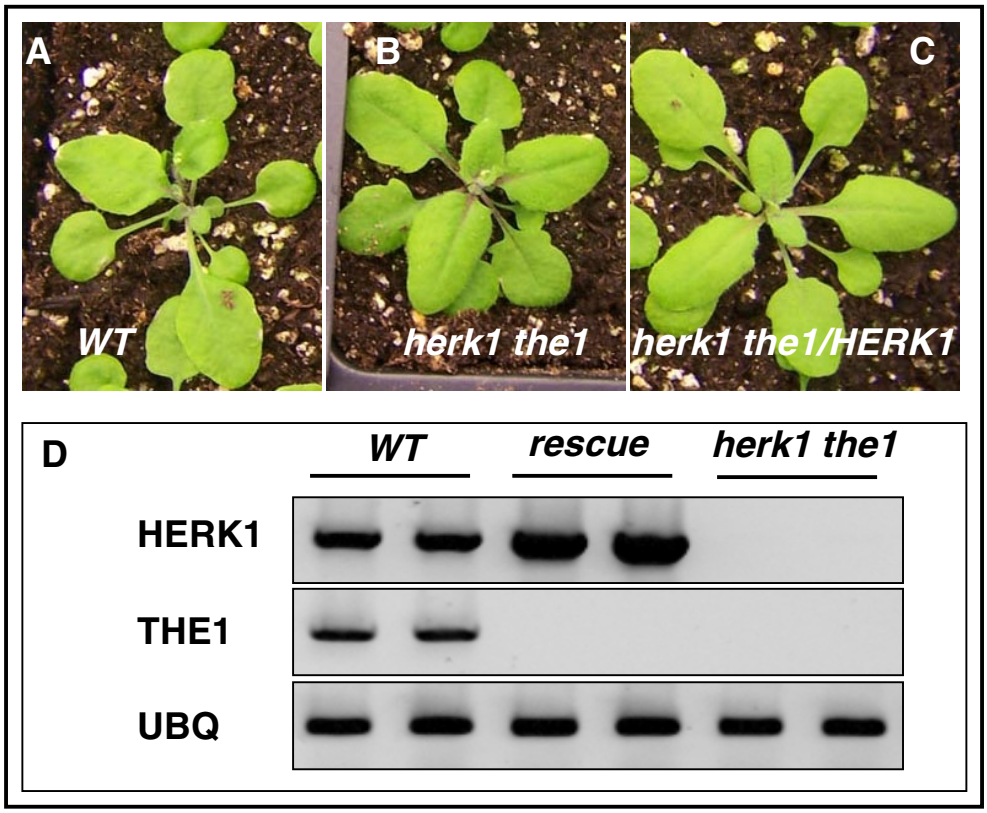

Fig. S2. The expression of HERK1 in *herk1 the1* mutant background rescued the mutant phenotype. A full-length genomic clone of HERK1 was transformed into the double mutant plants by floral-dip method. (*A*–*C*) Four-week-old wild-type (WT, A), *herk1 the1* mutant (*B*) and one of the representative rescued T2 plant lines (*C*) are shown. (*D*) RNAs prepared from these plant lines were used in RT-PCR to detect the expression of *HERK1* and *THE1*. *UBQ5* gene was used as a control.

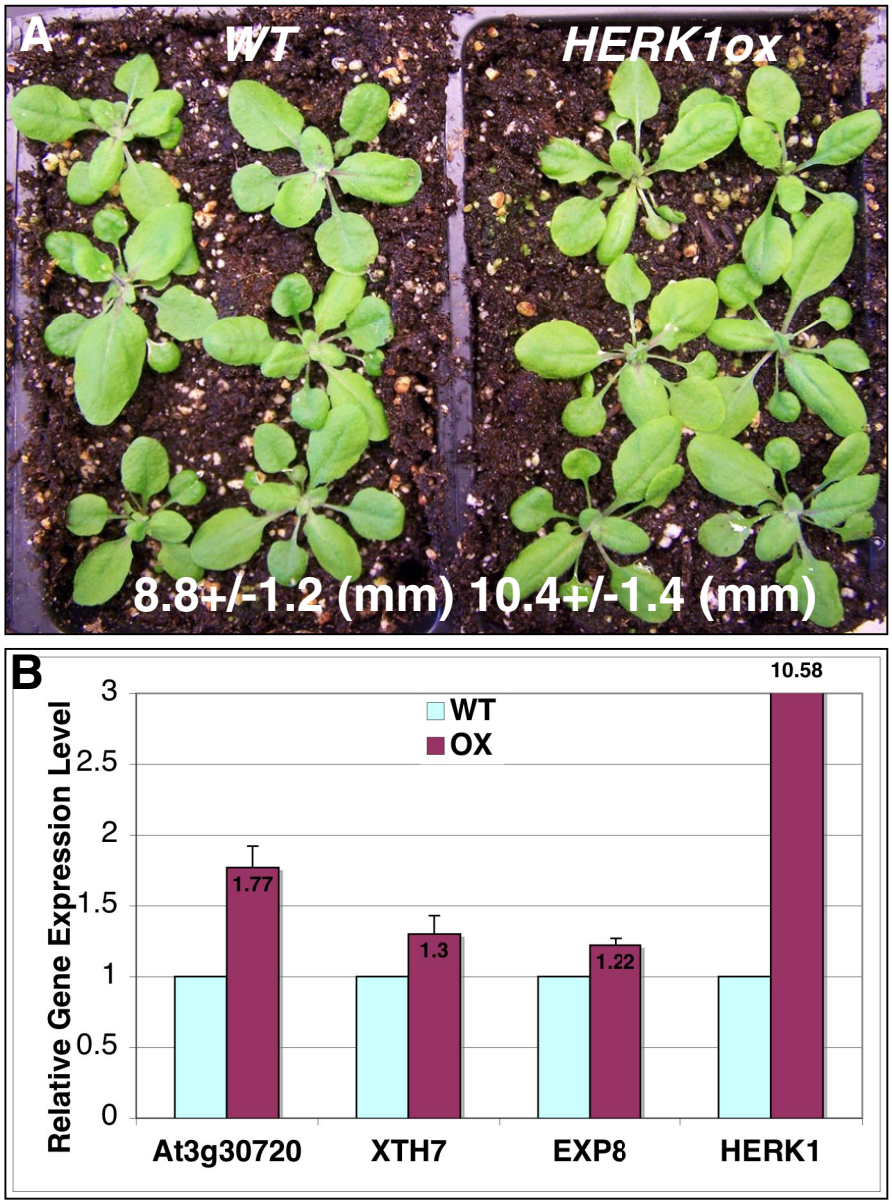

Fig. S3. HERK1 overexpression phenotype. (*A*) Shown are 22-day-old plants of WT and a HERK1 overexpression line (HERK1ox). The overexpression lines were identified from transgenic plants carrying the HERK1 genomic clone including the promoter. As indicated in the figure, overexpression transgenic plants have slightly increased petiole lengths compared to WT (average and standard deviations from 10 transgenic plants). The difference was significant according to student's *t* test ($P < 0.1$). (*B*) Several genes down-regulated in *herk1 the1* mutant (Fig. 4) are increased in *HERK1* overexpresson plants. The relatively subtle phenotype of the HERK1 overexpression lines (although the gene was overexpressed 10 times with its native promoter) suggests that other components in the HERK pathway (such as ligand, coreceptor and/or downstream signaling components) are likely more rate-limiting.

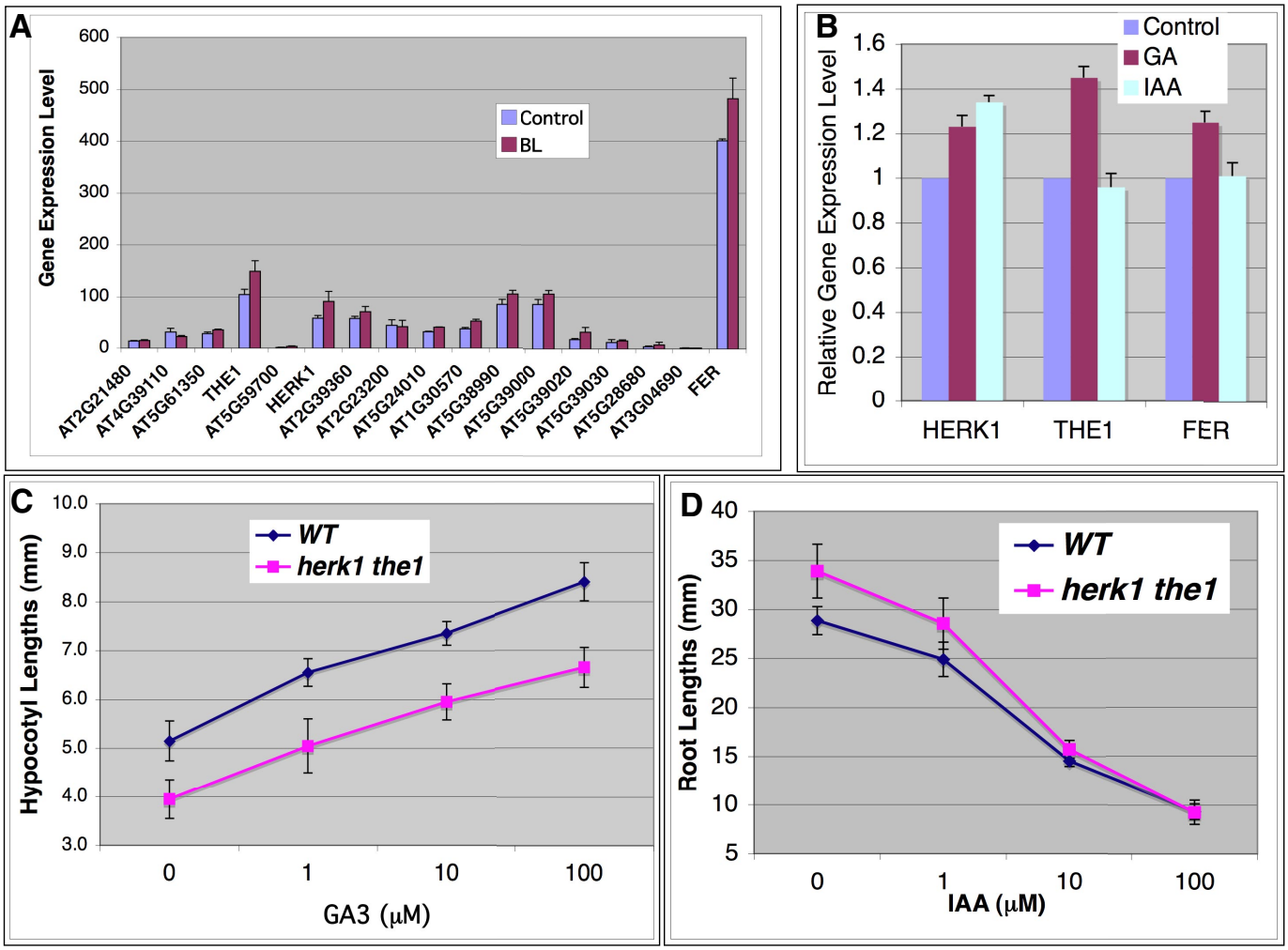

Fig. S4. Regulation of CrRLK genes and gibberellin (GA)/auxin responses of *herk1 the1* double mutant. (*A*) Regulation of *HERK1*, *THE1*, *FER* and other CrRLK family members by BL. The data are derived from microarray experiments with light-growing seedlings treated with 10 nM BL for 3 h, which is published in a public website (http://bbc.botany.utoronto.ca/efp/cgi-bin/efpWeb.cgi). (*B*) Regulation of *HERK1*, *THE1*, and *FER* by GA3 and auxin (IAA) in 10-day-old seedlings as revealed by qRT-PCR. The averages and standard deviations for 2 biological replicates are shown. (*C*) The *herk1 the1* double mutant has a normal GA response in a hypocotyl elongation assay. The seeds were germinated in 1/2MS media containing the indicated concentrations of GA3 and grown under the light for 10 days. (*D*) *herk1 the1* double mutants had a slightly increased response to auxin in root elongation assay. Seeds were germinated and grown in media with the indicated concentration of IAA in vertical plates for 11 days. The increased sensitivity to IAA may be due to reduced BR response in *herk1 the1* double mutant because BR loss-of-function mutants also display hypersensitivity to auxin in root elongation assays [Ephritikhine G, Fellner M, Vannini C, Lapous D, Barbier-Brygoo H (1999) The *sax* dwarf mutant of *Arabidopsis thaliana* shows altered sensitivity of growth responses to abscisic acid, auxin, gibberellins and ethylene and is partially rescued by exogenous brassinosteroid. *Plant J* 18:303–314.]. The averages and standard deviations are derived from 20–30 samples for both GA and IAA response experiments.

Other Supporting Information Files

Table S1
Table S2
Table S3
Table S4
Table S5