Α

100% 80% 60% 40% 20% 0% EPL1a (AT1G16690) -77% EPL1b (AT1G79020) -14% EPCR1 (AT4G32620) -20% EPCR2 (AT5G04670) -В EPCR1-Flag \rightarrow epcr1/2 epcr1/2 2 WT 1 1 cm 180 kDa αFlag αΑCTIN 48 kDa

Expanded View Figures

Figure EV1. Analysis of enhancer of polycomb-like proteins in Arabidopsis.

A Phylogenetic analysis of enhancer of polycomb-like proteins in Arabidopsis.

B The EPCR1-Flag transgene complements the developmental defects in the epcr1/2 mutant. We generated a construct harboring a native promoter-driven EPCR1 genomic sequence and transformed the construct into the epcr1/2 mutant for complementation testing. Two independent transgenic lines were selected for analysis. Morphological phenotypes of 10-day-old seedlings are shown. Western blot shows the expression of EPCR1-Flag protein in the epcr1/2 mutant, and ACTIN signals are shown as loading controls.

Source data are available online for this figure.

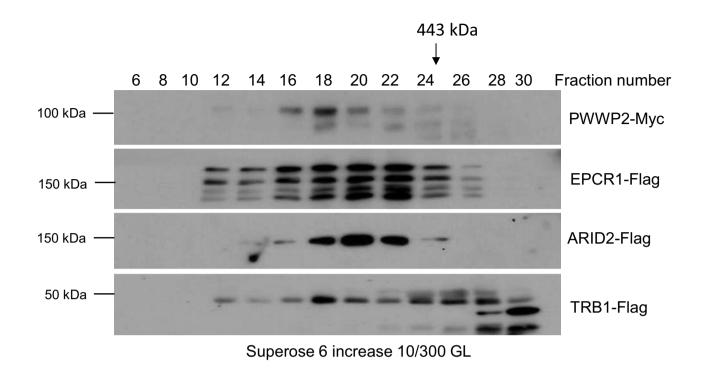


Figure EV2. PWWP, EPCR, ARID, and TRB proteins form complexes in vivo as determined by gel filtration.

Proteins were extracted from *PWWP2-Myc*, *EPCR1-Flag*, *ARID2-Flag*, and *TRB1-Flag* transgenic plants in the wild-type background. The proteins were eluted on a Superose 6 increase (10/300 GL) column. The epitope-tagged proteins in different fractions were detected by antibodies against the Myc or Flag-tag. Arrows indicate the fractions that correspond to the standard proteins 443 kDa.

Source data are available online for this figure.

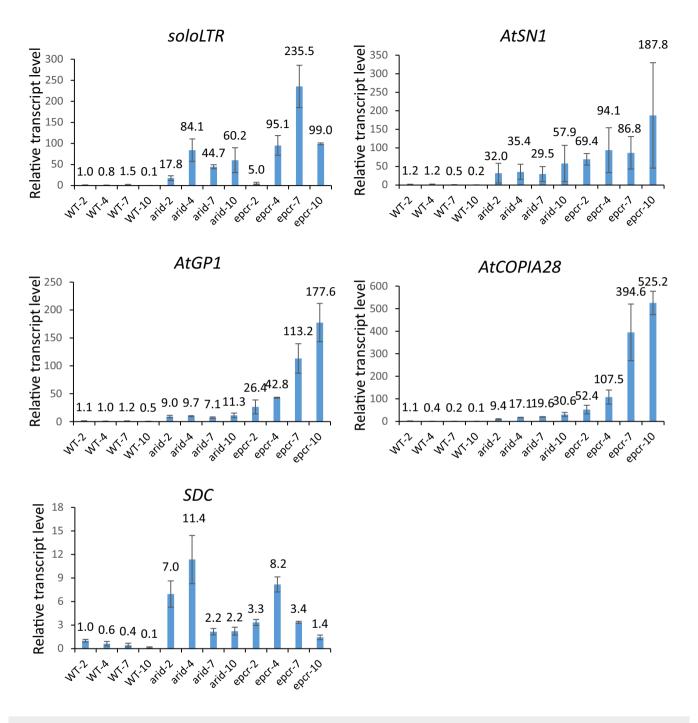


Figure EV3. Transcriptional silencing in the wild type and the *arid2/3/4* **and** *epcr1/2* **mutant seedlings at different development stages.** Transcript levels of *solo LTR, AtSN1, AtGP1, AtCOPIA28,* and *SDC* in the wild type and the *arid2/3/4,* and *epcr1/2* mutant seedlings grown for 2, 4, 7, and 10 days after germination. Shown are transcript levels of indicated loci, as determined by qPCR analysis. Error bars represent SD from three independent biological replicates.

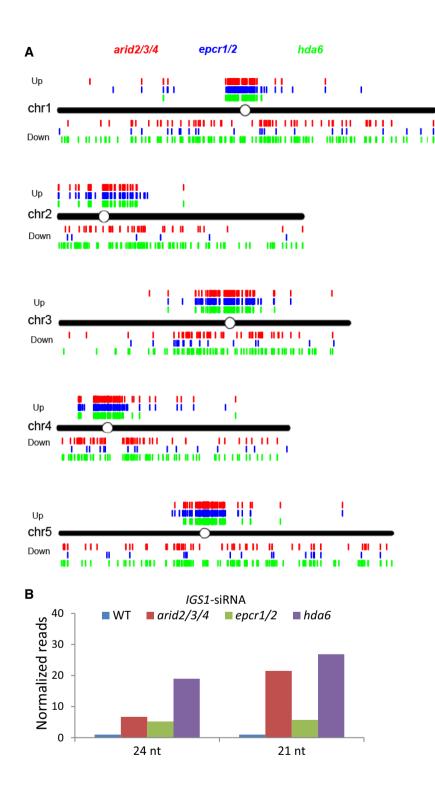


Figure EV4. Effect of the *arid2/3/4*, *epcr1/2*, and *hda6* mutations on small RNA accumulation.

- A Distribution of the Pol IV-dependent siRNA regions that are differentially expressed in the *arid2/3/4, epcr1/2,* and *hda6* mutants relative to the wild type. "Up" and "Down" refer to the Pol IV-dependent siRNA regions in which the siRNAs are up- and downregulated, respectively, in the indicated mutants relative to the wild type.
- B Relative levels of 21- and 24-nt siRNAs from the 45S rDNA intergenic spacer locus *IGS1* in the wild type, *arid2/3/4*, *epcr1/2*, and *hda6* mutants. The RNA levels were indicated by normalized small RNA reads as determined by our small RNA-seq analysis.

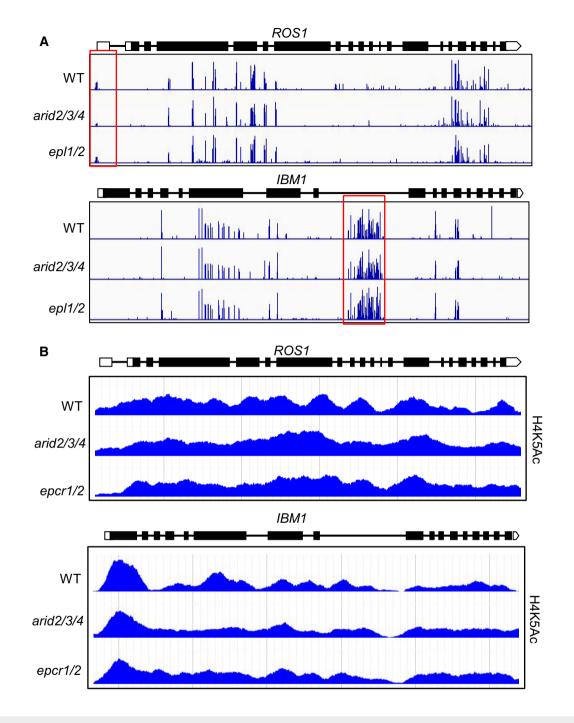


Figure EV5. Genome browser snapshots showing DNA methylation and histone H4K5 acetylation patterns of ROS1 and IBM1 in wild type, arid2/3/4, and epcr1/2.

- A DNA methylation of *ROS1* and *IBM1* as determined by the whole-genome DNA methylation analysis. The length of blue lines represents DNA methylation levels of cytosine sites. Red boxes highlight genomic loci harboring DNA methylation that is required maintenance of the expression of *ROS1* and *IBM1-L*.
- B Histone H4K5 acetylation of ROS1 and IBM1 as determined by the histone H4K5Ac ChIP-seq experiment.