

Genome-wide distribution and functions of the AAE complex in epigenetic regulation in *Arabidopsis*

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SUPPORTING INFORMATION

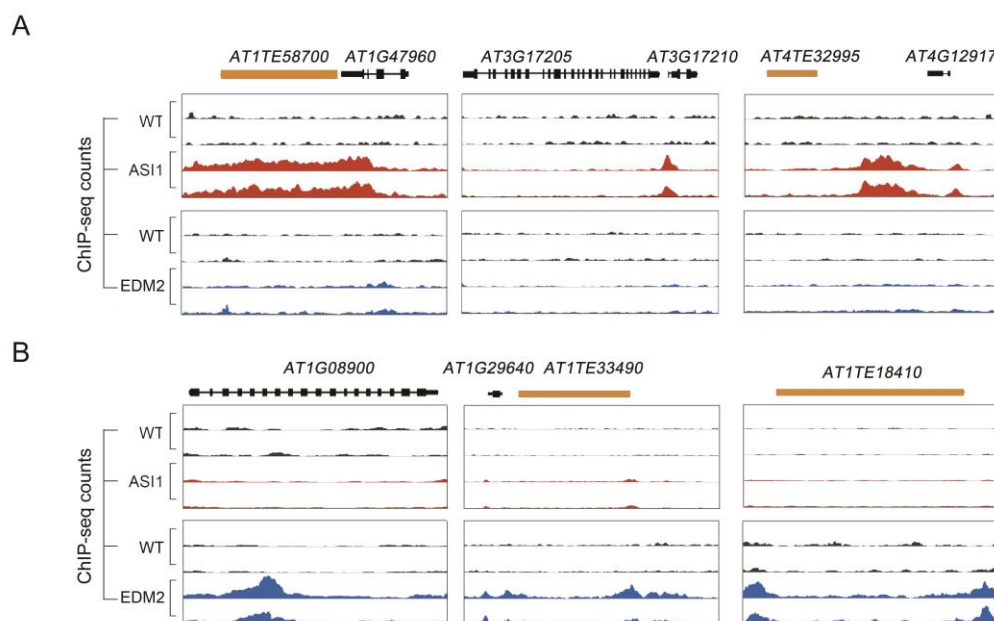
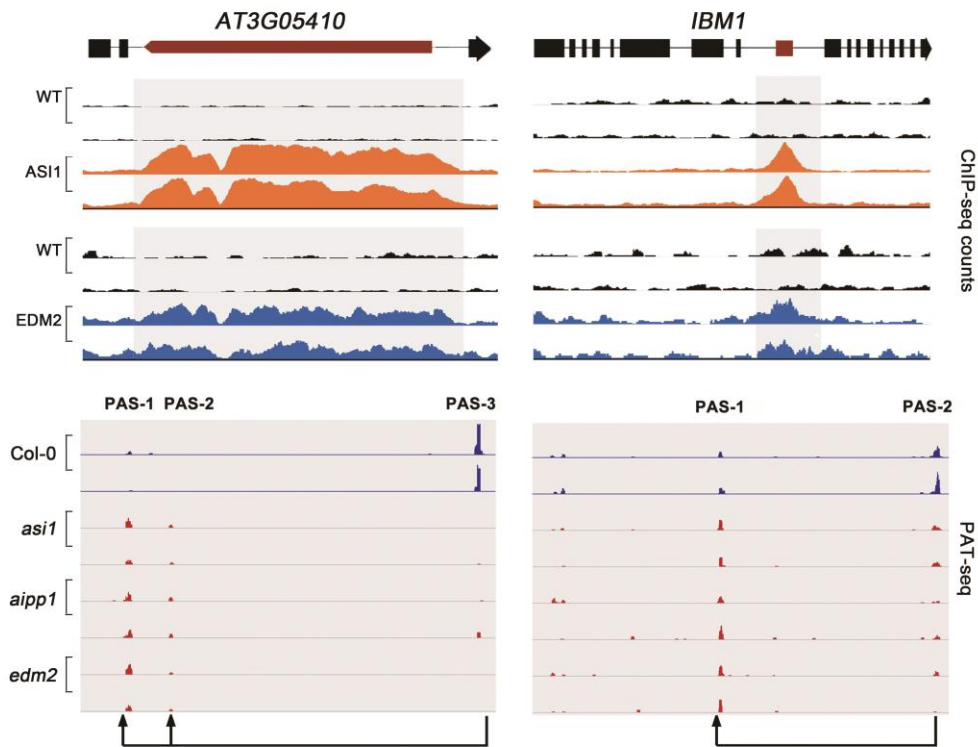


Figure S1. Representative genomic regions that are uniquely bound by ASI1 or EDM2

Snapshots of IGV from the ASI1 and EDM2 ChIP-seq analysis showing the genomic regions uniquely bound by ASI1 (A) or EDM2 (B). Orange bars represent TEs.

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25 **Figure S2.** AAE complex dysfunction does not affect the genome-wide profile
26 of poly(A) site usage

27 Upper panel: snapshots of IGV from the ASI1/EDM2 ChIP-seq and PAT-seq
28 analysis showing the binding regions (upper panel) of ASI1 and EDM2 at
29 intronic heterochromatin-containing genes *AT3G05410* and *IBM1*. Lower panel:
30 snapshots of IGV from PAT-seq indicating poly(A) site usage in Col-0 and the
31 selected mutants. Black and orange boxes represent exons and
32 heterochromatin regions (gray regions), respectively. The black arrows
33 indicated the switch of poly(A) usage from distal sites to proximal sites.

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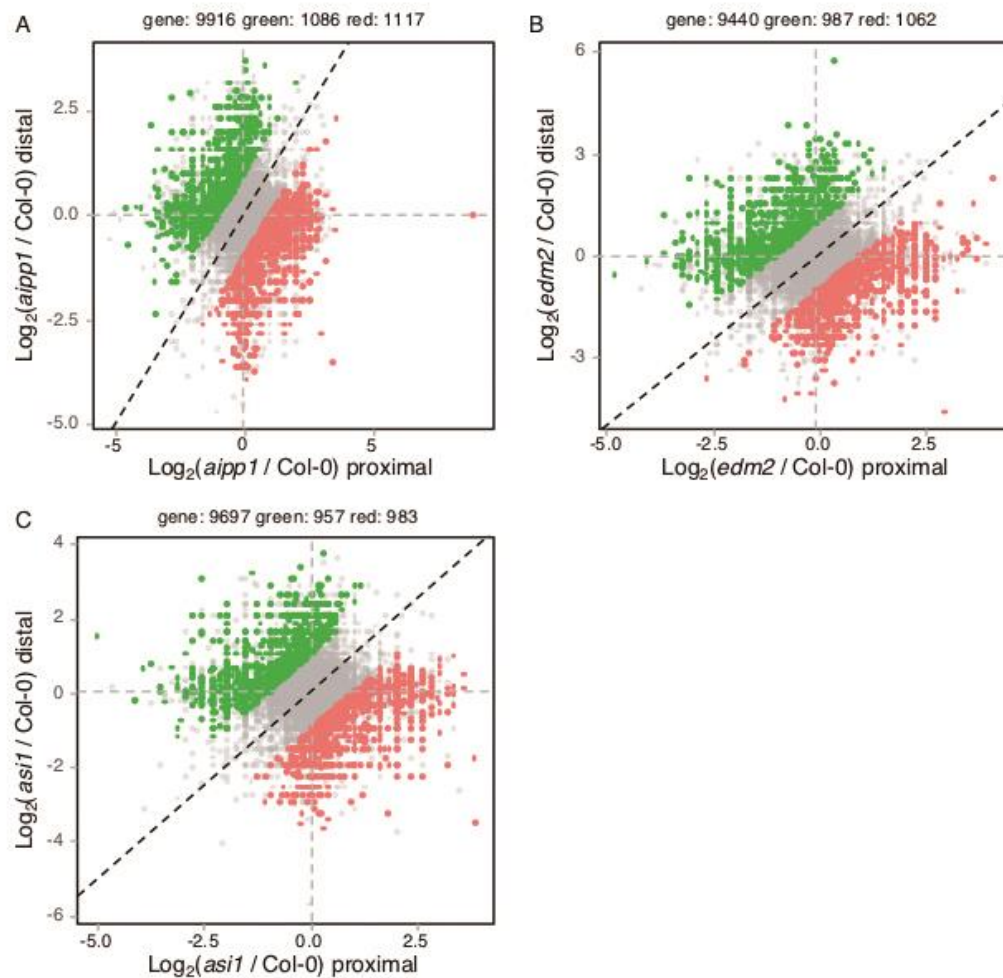
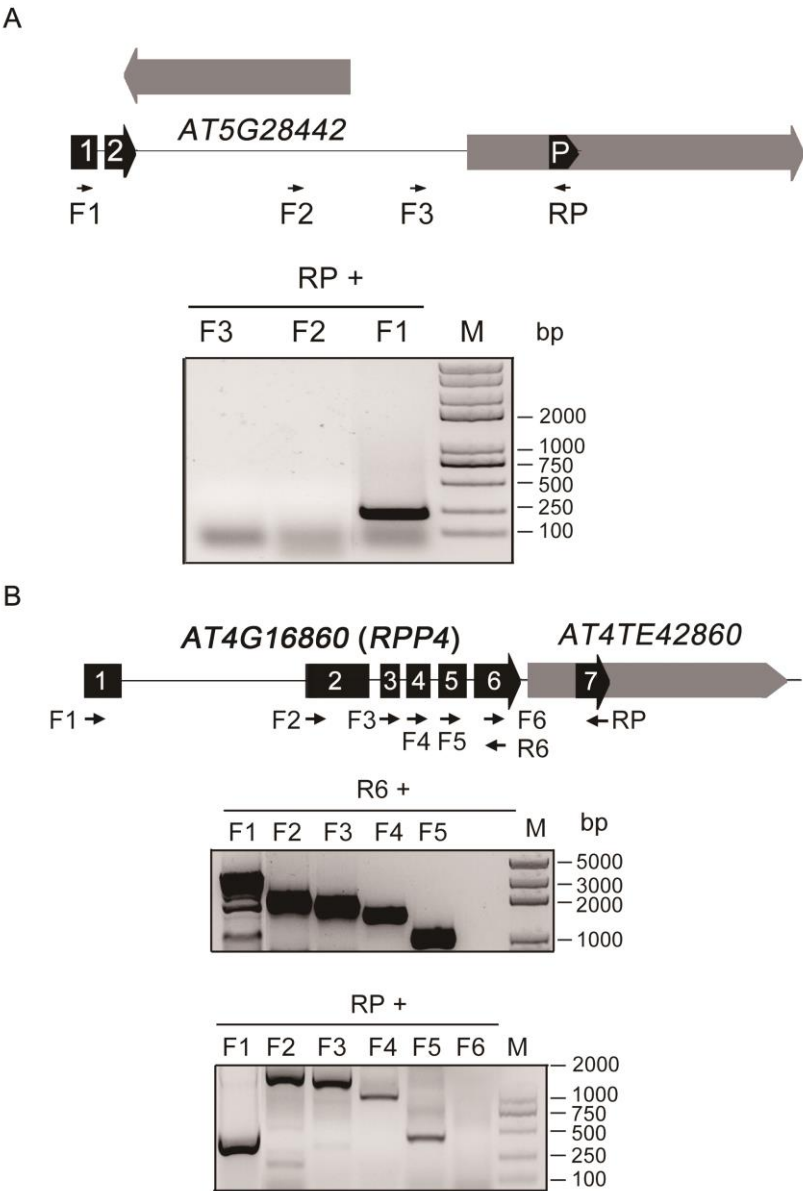


Figure S3. Alternative polyadenylation events in *aipp1*, *edm2* and *asi1* mutants

Top two expressed poly(A) sites were selected for analysis. Diagonal lines indicate theoretical no shifting APA between mutants and Col-0. Grey dots represent no significant shift. Green dots indicate significant shift from proximal poly(A) sites to distal poly(A) sites. Red dots indicate significant shift from distal poly(A) sites to proximal poly(A) sites



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54 **Figure S4.** Alternative splicing of the intragenic heterochromatin-containing
55 *RPP4* and *AT5G28442* genes

56 RT-PCR results from agarose gel staining show the alternative transcripts
57 generated by the *AT5G28442* (**A**) and *RPP4* (**B**) genes. RT-PCR was
58 performed using total RNAs from Col-0 wild type plants. The primers used for
59 PCR amplification are labeled. Black and gray boxes represent exons and TE,
60 respectively. P, predicted exon. M, DNA marker.

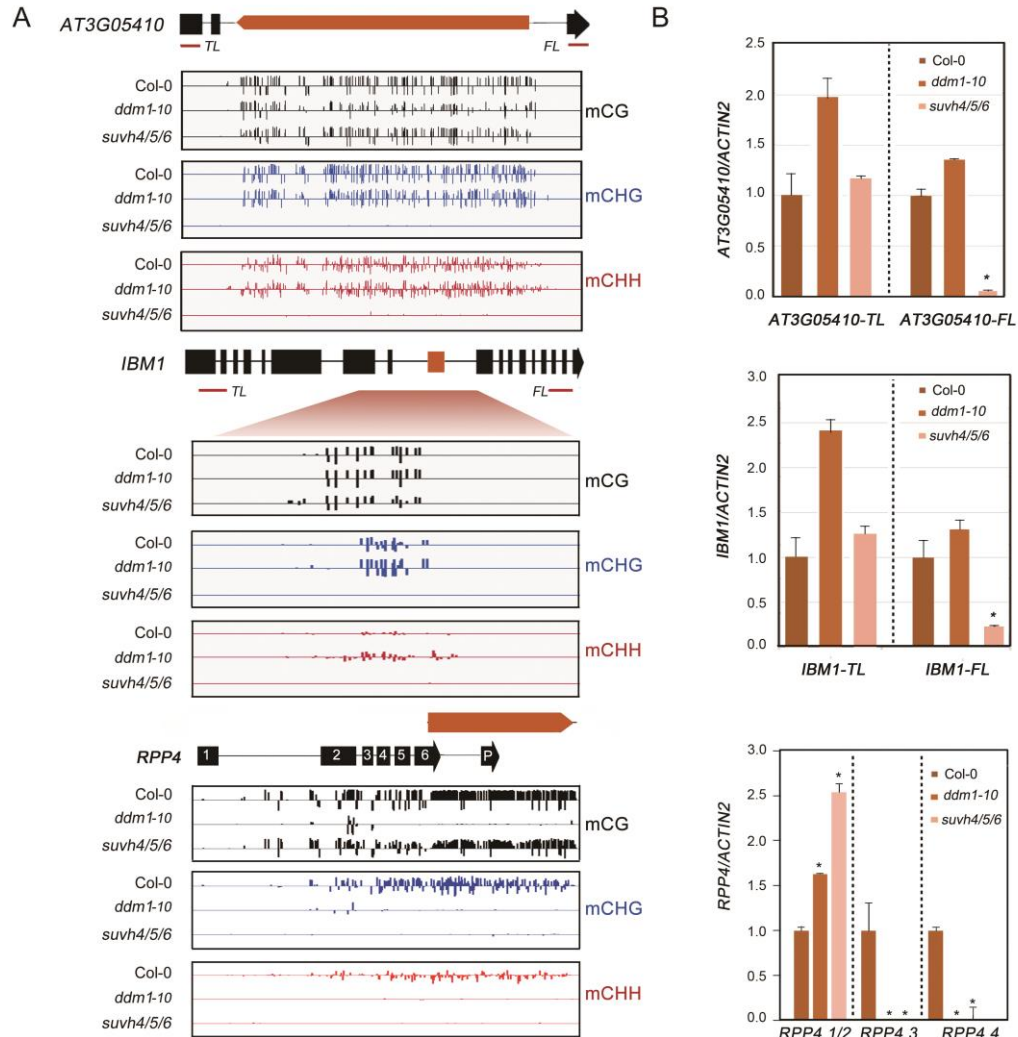


Figure S5. The mis-expression of selected AAE complex target genes in the *ddm1* and *suvh4/5/6* mutants

(A) IGV snapshots showing the DNA methylation levels of selected AAE complex target genes in Col-0, *ddm1-10* and *suvh4/5/6* mutants. DNA methylation levels were calculated according to published DNA methylome data of Col-0 plants. The black, blue and red lines represent CG, CHG and CHH methylation, respectively. The black and orange boxes indicate exons and TEs. (B) RT-qPCR showing the relative expression of the alternative transcripts. Primer pairs corresponding to the upstream and downstream

exons of the intronic TEs were designed to monitor the expression levels of full-length (*L*) and total (*TL*) transcripts. The positions are labeled in A. Relative expression levels were first normalized to the expression level of *ACTIN2* and then to that of the wild-type plants. Error bars indicate *SD*; N=3. **p*<0.01, Student's test in comparison with Col-0.

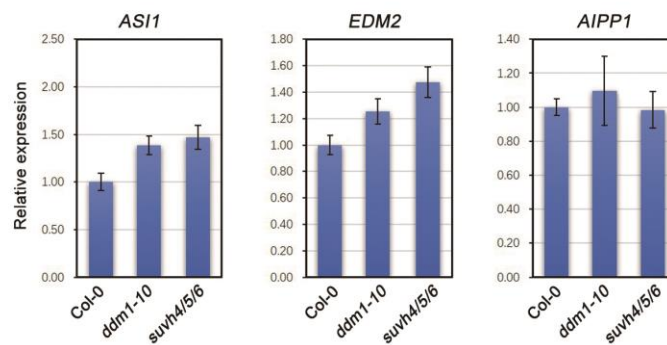
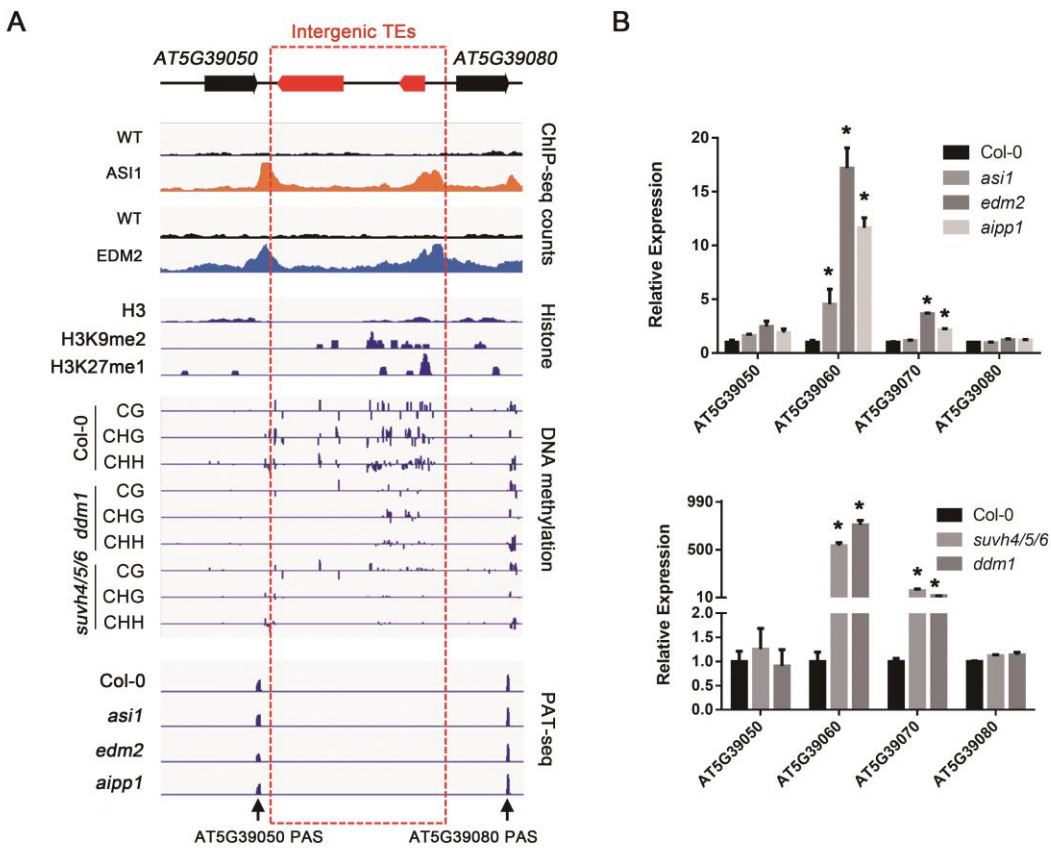


Figure S6. The relative expression of *ASI1*, *EDM2* and *AIPP1* genes in the *ddm1* and *suvh4/5/6* mutants

The expression levels of the *ASI1*, *EDM2* and *AIPP1* genes were examined in different mutants. Relative expression levels were first normalized to *ACTIN2* and then to those of the wild-type plants. Error bars indicate *SD*; N=3.



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100 **Figure S7.** The effects of AAE complex dysfunction on the expression
101 intergenic TEs and adjacent genes

102 **(A)** IGV snapshots showing the distributions of ASI1/EDM2 ChIP-seq counts,
103 heterochromatic histone marks, DNA methylation levels in different genotypes
104 as well as the poly(A) site usage in the intergenic TEs and adjacent genes.
105 Red dashed box indicates the heterochromatic regions bound by ASI1 and
106 EDM2. Red and black boxes represent TEs and exons, respectively. **(B)**
107 RT-qPCR results showing the relative expression of TEs and adjacent genes
108 in different genotypes. *, p value<0.01, Student's test.

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110 **Table S1.** ASI1 and EDM2 ChIP peaks

111 **Table S2.** A-E-bound genes

112 **Table S3.** Primers