Supporting Information

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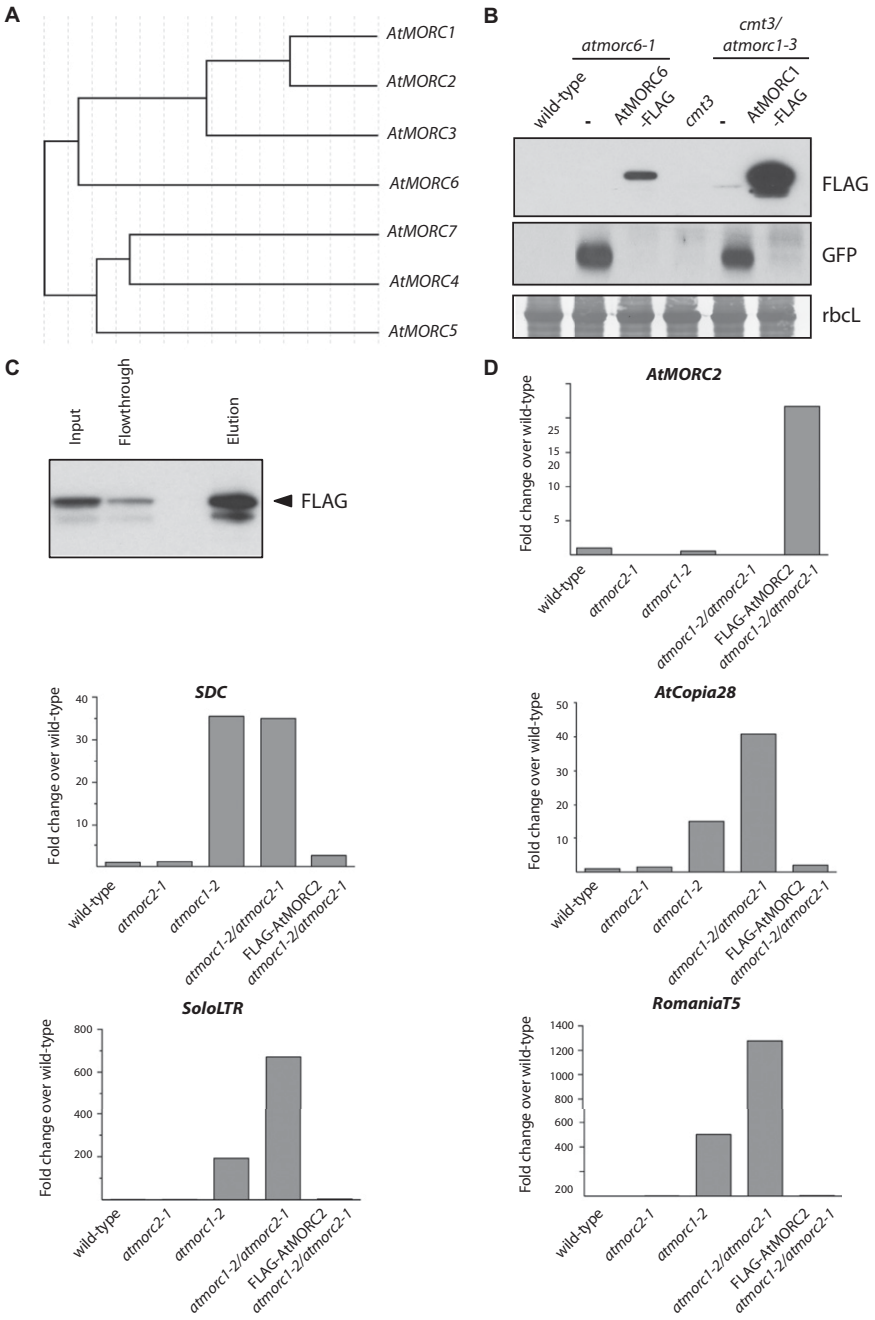

Fig. S1. Phylogenetic analysis and epitope-tagging of *Arabidopsis* Microrchidia (AtMORC)1, AtMORC2, and AtMORC6. (*A*) Phylogenetic analysis of the *AtMORC* gene family in *Arabidopsis thaliana*. The alignment was made with Genebee using the default parameters. (*B*) AtMORC1 and AtMORC6 transgenic lines. FLAG epitope-tagged *AtMORC1* and *AtMORC6* were expressed under their respective endogenous promoter in their respective mutant background. Protein expression and complementation of the *SDC::GFP* silencing defects were probed by Western blotting. The large subunit of rubisco (rbcL) was used as the loading control. (*C*) Transgenic AtMORC2 line. FLAG epitope-tagged AtMORC2 was expressed under its respective endogenous promoter in the *atmorc1/atmorc2* double mutant background. FLAG-AtMORC2 is enriched in the elution fraction after immunoprecipitation. (*D*) Complementation of transcriptional derepression by expression of FLAG-AtMORC2 in *atmorc1/atmorc2*. RT-PCR shows increased levels of FLAG-AtMORC2 transcripts compared with wild-type and *atmorc1-2* despite being expressed under its respective endogenous promoter. Derepression of *suppressor of drm2 cmt3* (*SDC*)*::GFP*, *AtCopia28*, *Solo long terminal repeat* (*LTR*), and *RomaniaT5* is suppressed by overexpression of FLAG-AtMORC2.

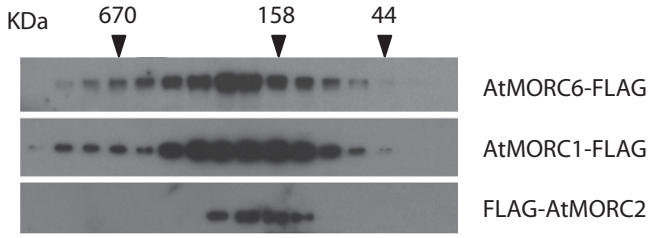

Fig. S2. AtMORC1, AtMORC2, and AtMORC6 copurify by gel filtration. AtMORC6-FLAG, AtMORC1-FLAG, and FLAG-AtMORC2 leaf protein extracts were separated on a Superdex 200 10/300GL column. Eluting fractions were immunoblotted with antibodies directed against FLAG. Sizing standards are shown.

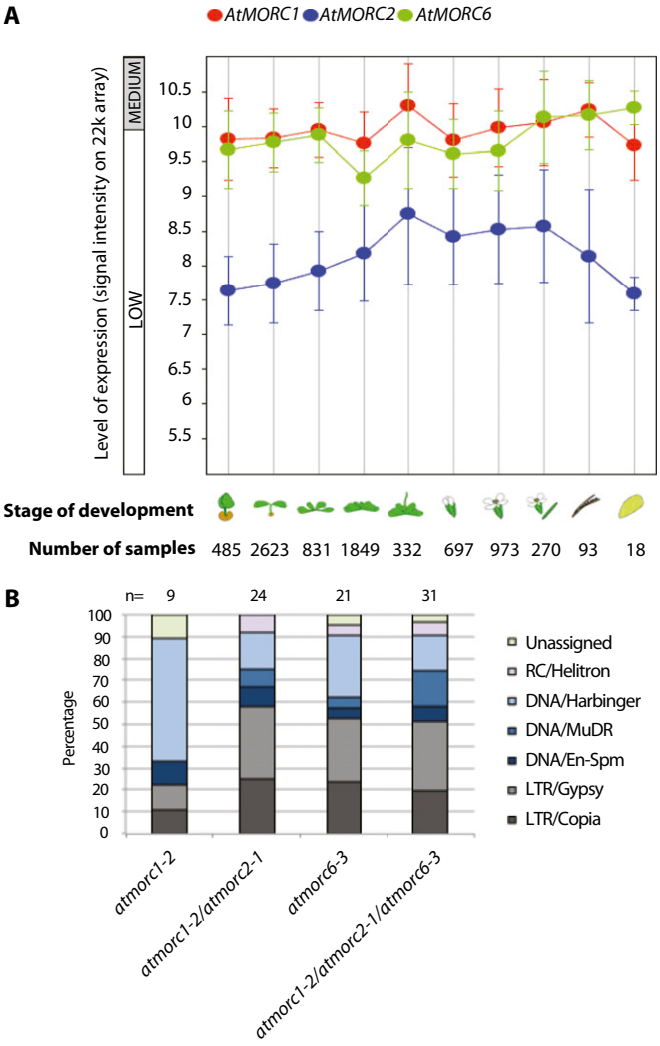

Fig. S3. Analysis of *AtMORCs*' expression and derepressed transposon families in *atmorc* mutants. (*A*) Transcript levels of *AtMORC1*, *AtMORC2*, and *AtMORC6* over stages of development. Gene expression data were retrieved from Genevestigator (www.genevestigator.com). (*B*) Transposons significantly derepressed in the different genotypes grouped according to their family.

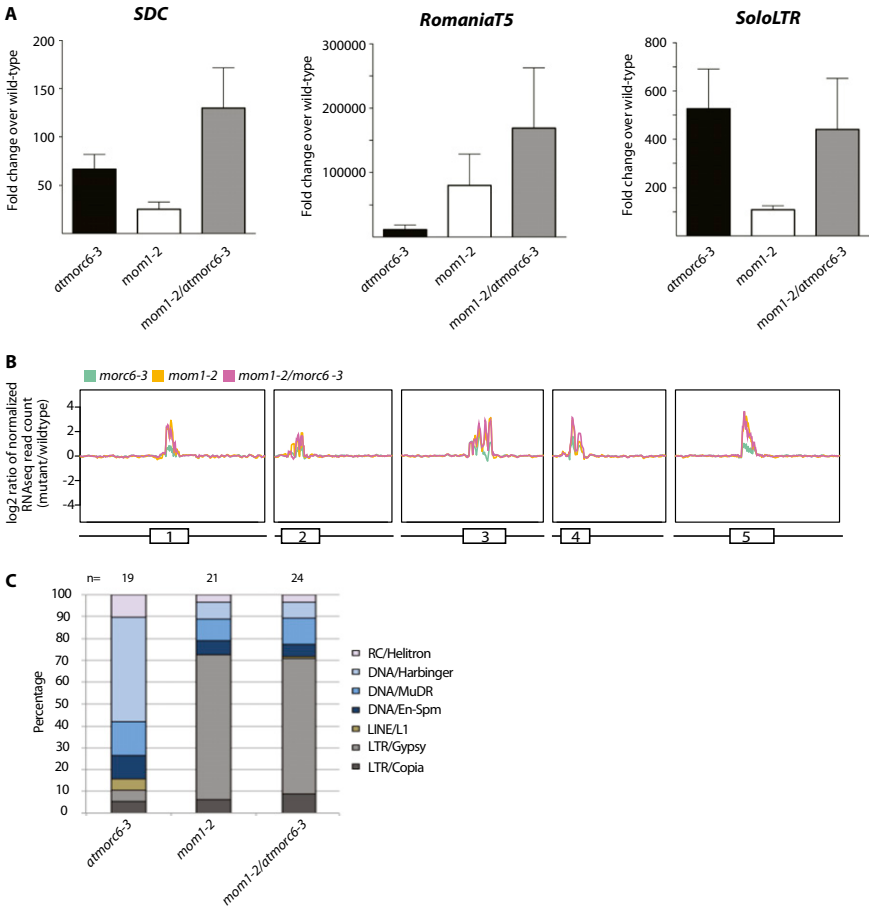

Fig. S4. Synergy of *AtMORC6* and *Morpheus Molecule 1* (*MOM1*) in transposon silencing. (*A*) RT-PCR assessing endogenous expression of *SDC*, *RomaniaT5*, and *SoloLTR*. Two biological replicates were performed for each tested genotype. (*B*) Chromosomal view of percent change in read abundance between higher order mutants and other genotypes. (*C*) Transposons significantly derepressed in the different genotypes grouped according to their family.

Table S1. Primers used in this study

Genotyping

Genotype	Insertion line	Left border primer	Right border primer
atmorc1-2	SAIL_893_B06	TTGCAGTTTGGAACCAAAATC	TGAGTTTTGACGACGATGATG
atmorc1-4	SAIL_1239_C08	CGTATCTCAGCCGCTAACTTG	AAGCAGCTGCAGTGGATTATG
atmorc1-5	SAIL_131_H11	CGTATCTCAGCCGCTAACTTG	AAGCAGCTGCAGTGGATTATG
atmorc2-1	SALK_072774C	CTACTCAGAGCGTTGGCATTC	GTTGTAGCTGTATGGGGCTTG
atmorc2-4	SALK_021267C	TTTCGTCATCATTGCTTTTCC	GGTTGACTCTTCCACTGCTTG
atmorc6-3	GABI_599B06	GGAAAGCTGGAAGCTATAATGATG	GATGACATCTGCCCCAAGTCTC
cmt3-11	SALK_148381	TAACGGAAGGATGCCAGATT	CAAGAAATGGGCTGTTGACAT
mom1-2	SAIL_610_G01	ACAATGCAGGAGCAAACACTC	GGAAAGGAGATACTTCACCGG
LB3	SAIL T-DNA	TAGCATCTGAATTTCATAACCAATCTCGATACAC	—
LBA1	SALK T-DNA	TGGTTCACGTAGTGGGCCATCG	—
LB O8409	GABI-KAT	ATATTGACCATCATACTCATTGC	—

RT-PCR

Locus	Left border primer	Right border primer
Actin 7	TCGTGGTGGTGAGTTTGTTAC	CAGCATCATCACAAGCATCC
SDC	AATGTAAGTTGTAAACCATTTGAACGTGACC	CAGGCATCCGTAGAACTCATGAGC
AtCopia28	AGTCCTTTTGGTTGCTGAACA	CCGGATGTAGCAACATTCACT
RomaniaT5	GTATCCTTTGGCCCGGTATT	GCCTCTTCGAAATGCCATAA
SoloLTR	AACTAACGTCATTACATACACATCTTG	AATTAGGATCTTGTTTGCCAGCTA