

Expanded View Figures

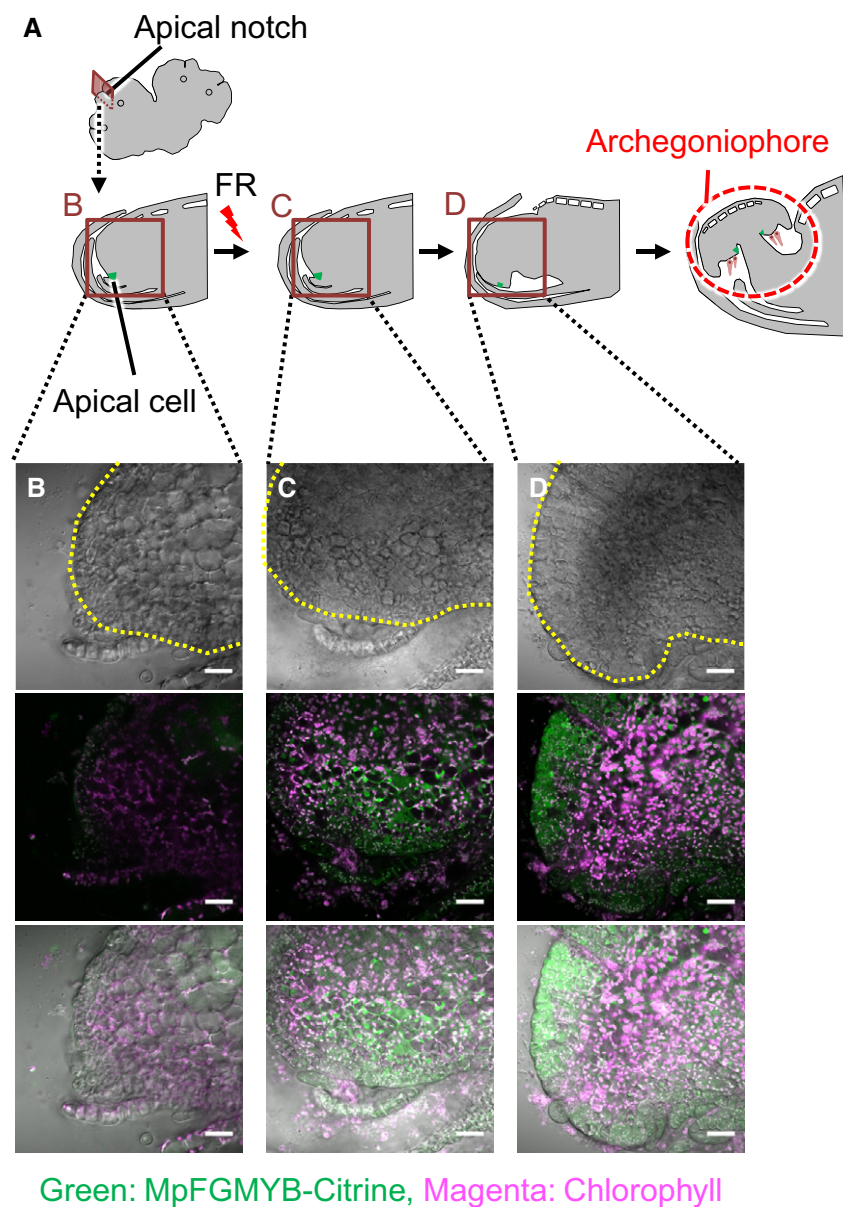


Figure EV1. MpFGMYB expression precedes archegoniophore morphogenesis.

- A** A schematic representation of the process of archegoniophore morphogenesis after far-red irradiation. Longitudinal sections of apical notch regions are presented. Around 14 days after induction, dome-shaped archegoniophore primordia developed at the ventral side of apical notch area. Regions corresponding to the images shown in (B–D) are boxed.
- B–D** Confocal microscopic images of the apical notch region of *gMpFGMYBresist-Citrine* plants without (B) or with far-red irradiation (C, D). MpFGMYB-Citrine does not accumulate in the apical notch region of vegetative thalli (B). After 10 days of FR irradiation, MpFGMYB-Citrine accumulates in the ventral side of the apical notch region (C). Expression domain of MpFGMYB-Citrine expands when the morphology of archegoniophore primordia becomes evident (D). Yellow dotted lines delineate the edges of thalli and a developing archegoniophore. Top, bright field images; middle, fluorescent images; bottom, merged images. Scale bar, 25 μ m.

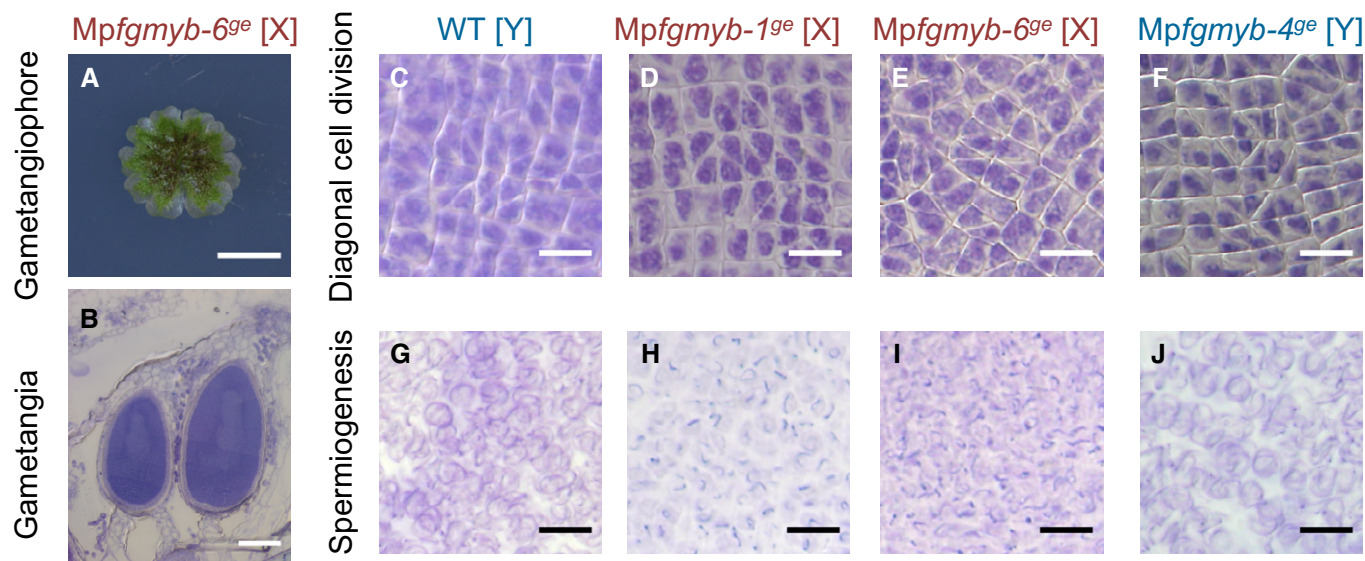


Figure EV2. Multiple *Mpfgmyb* [X] alleles consistently exhibit the female-to-male sex conversion phenotype.

A Gross morphology of antheridiophores developed in *Mpfgmyb-6ge* [X].

B–J Histological analyses indicating antheridium formation (B), diagonal cell division of spermatogenous cells (C–F), and subsequent spermiogenesis (G–J) in the wild-type (C, G), two independent *Mpfgmyb* [X] mutants (D, E, H, and I), and one *Mpfgmyb* [Y] mutant (F, J).

Data information: Scale bars, 5 mm (A), 100 μ m (B), 10 μ m (C–J).

Figure EV3. Characterization of the sex conversion phenotypes of *Mpfgmyb* [X].

A RT–PCR analysis demonstrating loss of expression of female-specific autosomal genes in the antheridiophores of *Mpfgmyb* [X]. Note that X chromosome-linked genes are still expressed in *Mpfgmyb* [X] antheridiophores as in the wild-type females, despite their male-like sexual morphologies. Biological duplicates were analyzed for each genotype.

B–D Hoechst-stained wild-type archegonia treated with sperm from wild-type (B), *Mpfgmyb* [X] (C), and *Mpfgmyb* [Y] (D) plants, indicating the inability of *Mpfgmyb* [X] sperm to enter wild-type archegonia. Arrowheads, sperm in archegonial cavity. Dotted lines, egg cells. Scale bars, 10 μ m (B'–D', B''–D''), 50 μ m (B–D).

Source data are available online for this figure.

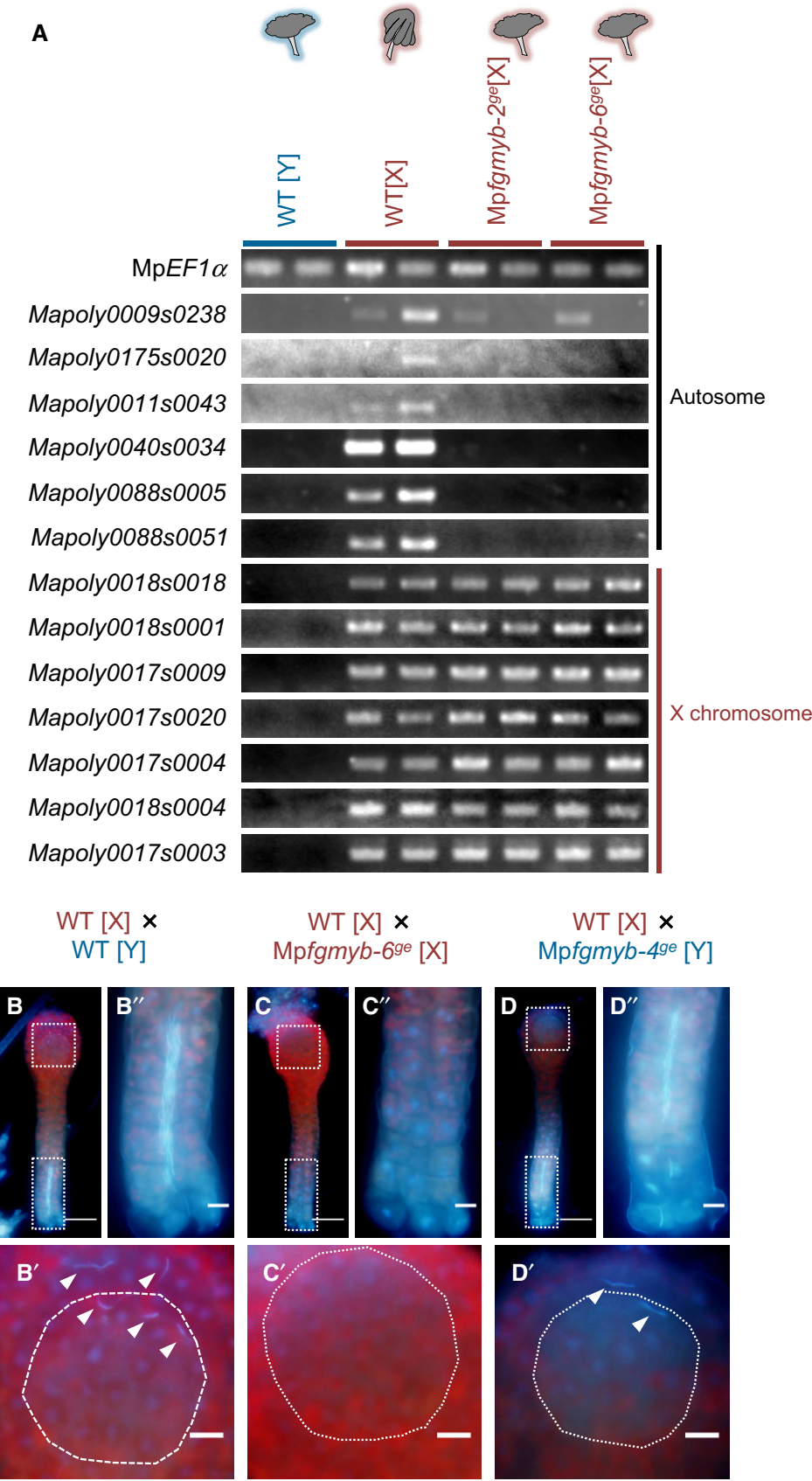


Figure EV3.

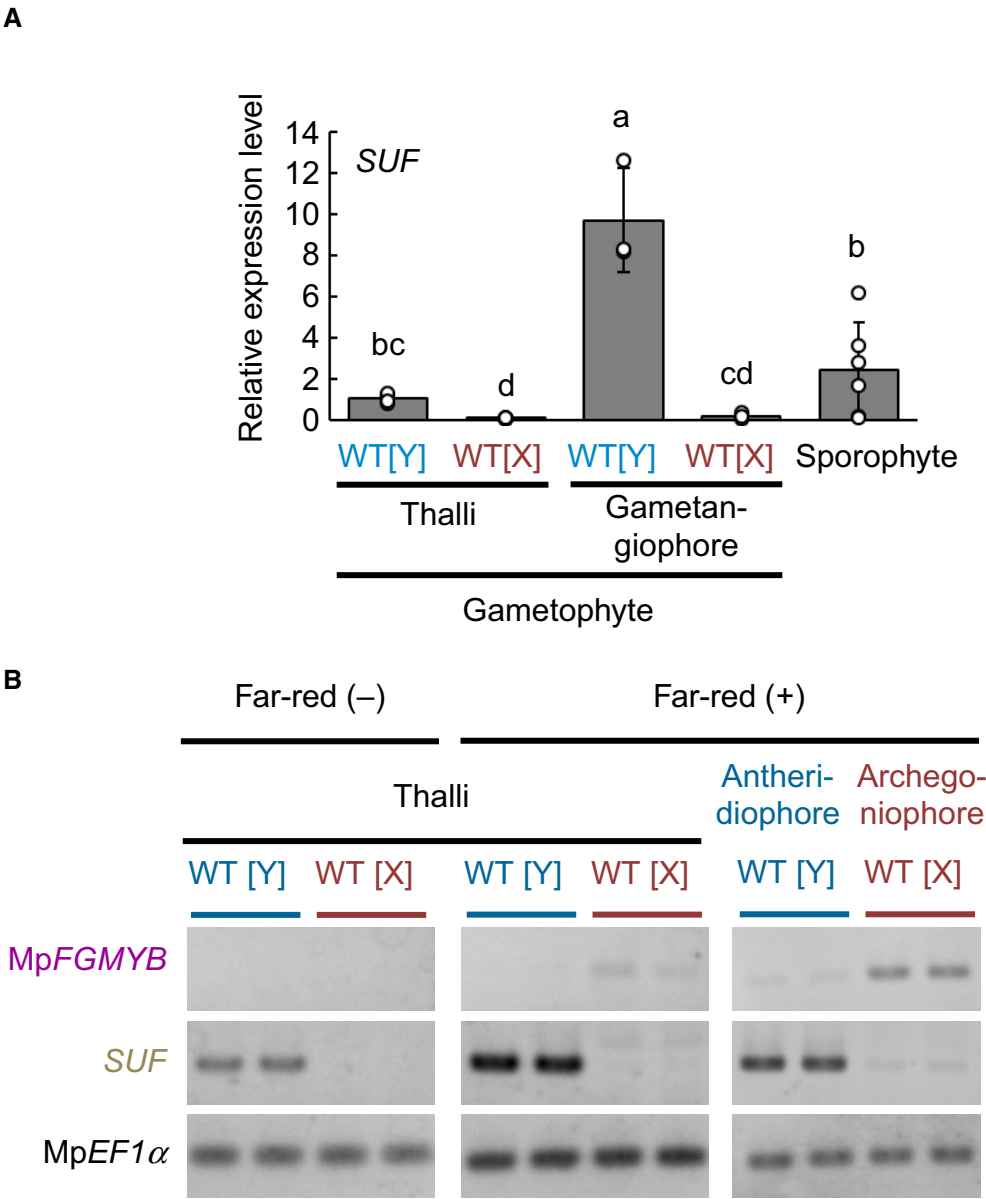


Figure EV4. Expression analyses of *SUF*.

A Real-time RT–PCR analyses indicating preferential accumulation of *SUF* transcripts in male reproductive organs. Constitutively expressed *MpEF1α* was used for normalization. Measurements of six biological replicates for thalli and sporophyte, and three biological replicates for gametangiophores are plotted. Bars represent mean $\pm$ SD. Symbols above bars indicate grouping by $P < 0.05$ in a Tukey–Kramer test. See Source Data online for measurements and statistics.

B Strand-specific RT–PCR confirmed male-specific accumulation of *SUF* transcripts in vegetative and reproductive organs, regardless of the induction of reproductive growth by far-red irradiation. Biological duplicates were analyzed for each sex. *MpEF1α* was used as a control.

Source data are available online for this figure.

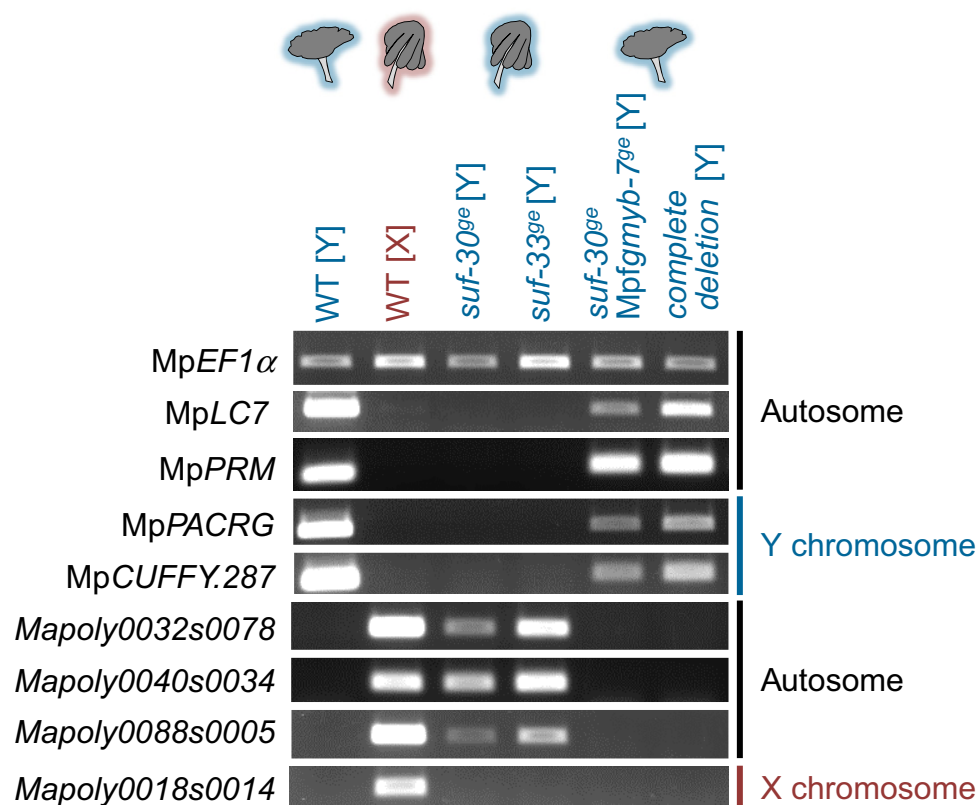


Figure EV5. Expression analyses of sex-specific genes in *suf* [Y].

RT-PCR analyses showing feminized gene expression patterns of *suf* [Y]. Note that the expression of female-specific genes in *suf* [Y] was suppressed by an additional mutation in the *MpFGMYB*-coding region. *MpEF1 α* was used as a control.

Source data are available online for this figure.