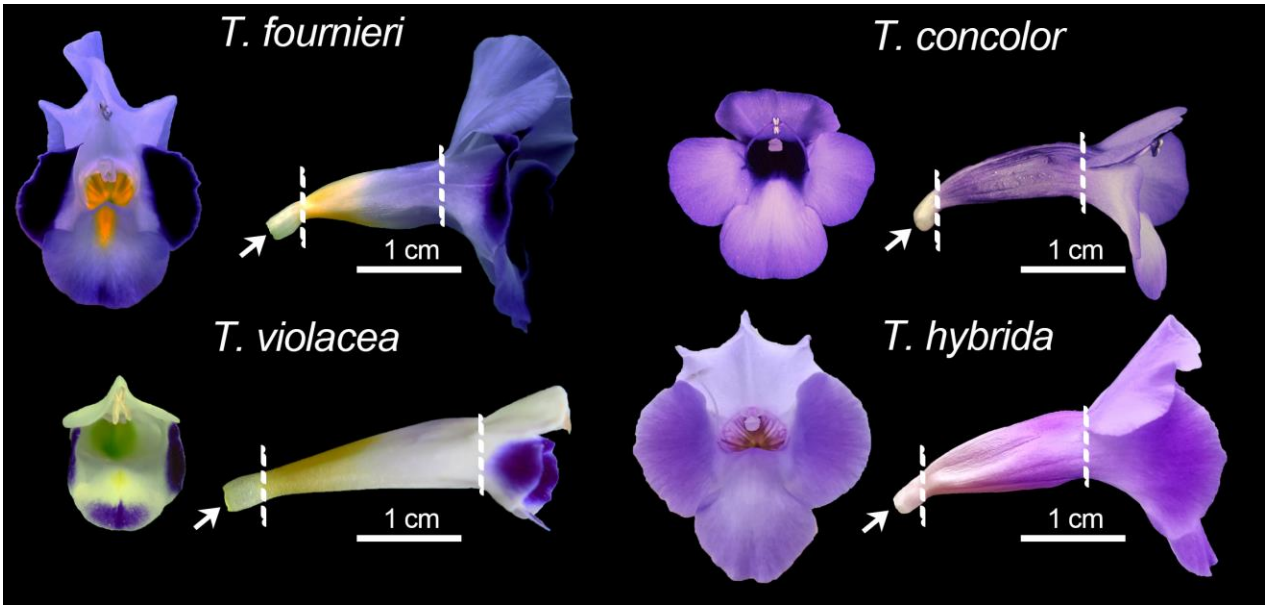

Fig. S1. Corolla morphologies of four *Torenia* species. Front and side views of each flower are shown. The dotted lines separate three different regions of the corolla, and the white arrows indicate the corolla neck.

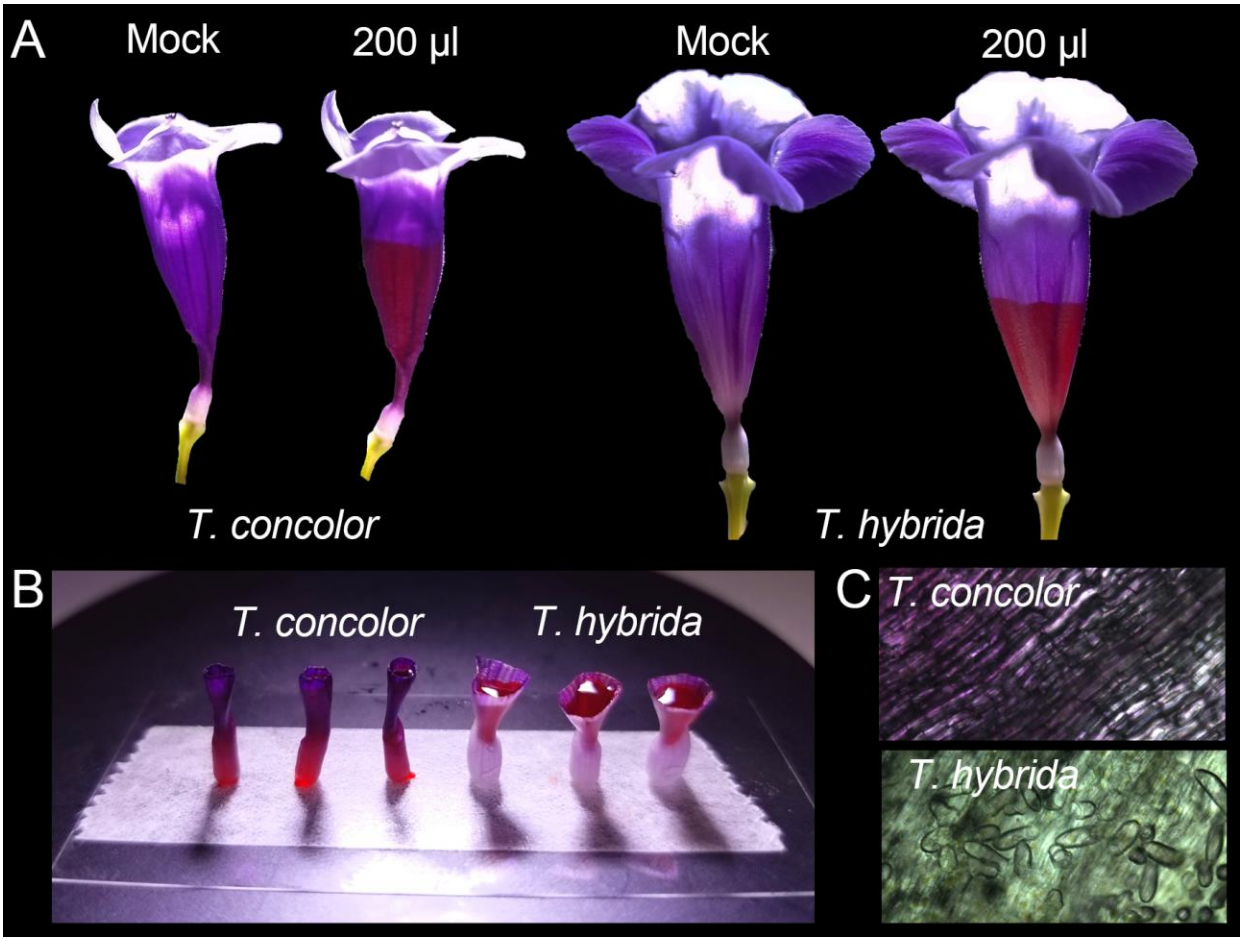

Fig. S2. Function and epidermal morphology of the corolla neck in *Torenia concolor* and *Torenia hybrida*. (A) Application of 200 µl water containing 0.1% Safranin O to a mature flower. (B) Application of 20 µl water containing 0.1% Safranin O to cut corolla bottoms. (C) Epidermal morphology of the corolla neck in *T. concolor* and *T. hybrida*.

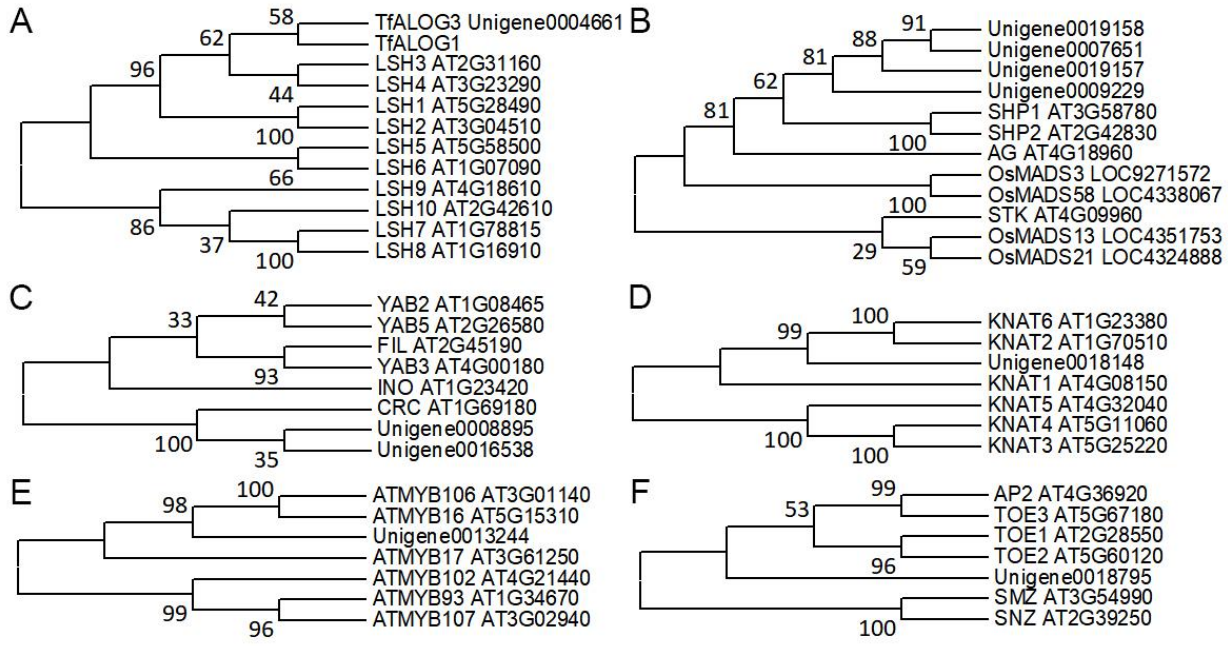

Fig. S3. Neighbor-joining trees for the top 10 enriched transcription factors. (A–F) The phylogenies of ALOG (A), MADS (B), YABBY (C), KNOX (D), MYB (E), and AP2 (F). The bootstrap value (percentage, based on 1000) is marked at each node, and the accession numbers are indicated as the suffix of each sequence.

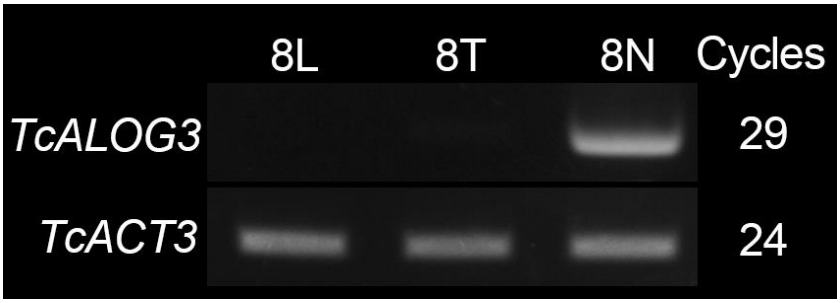

Fig. S4. Reverse-transcription PCR analysis in *Torenia concolor*. Gene names and number of PCR cycles are shown on the left and right sides of each panel, respectively. β-actin (*TcACT3*) expression was measured as an internal control. 8, 8 mm floral bud; L, petal lobe; T, conical petal tube; N, corolla neck.

A >TfALOG3 genomic DNA

ATGGACCCATCATTGTTCCCCAATCCTCAAAACGAGCCCACAAGCTCCGAGACCACCGCCAAC
ACCTACTCTGCAGCGACATCGTCGTCCTCCTCCACCTCCACCACCACCACCGCCGGCACATCCT
CCTCTTCCCGGTACGAGAACCAGAAACGCCGCGACTGGAACACCTTCAATCAGTATCTCAAGA
ACCACCGCCCGCCTCTCTCCCTCCACCGCTGCAGCGGGGCCCACGTCCTCGAATTTCTAAGGT
AGTAAGTAGTACTGCACCACTTGAACAAGAAACCTTTAAGTTTAATTGTTAAGACAGGGTGAG
TCGTGAGCAAGAATATGATCAAAGCGTTGAACGCGTTACTGTCGATAACAACTGATGTTGTAA
CGCAAACAAGTACGAGCAAAAGCTACACTGATAATATCTTATAACTCATAATTGGCCATCACAA
CATATCCCTAATTTACCTTATTTAATAAACGGCTGAGTTTCTCCCTCAATTCGGAGCTATTATTTT
CATCACGTTGTTGTTTTCGAGACTATCTTACTGCTGAACTACAGTTATTACATACGTGTTTGTTT
GTGTGTGTGTATATGTATGTATATATCTACCTCGCTATATCGTATAAACAGGTACCTTGACCAGTT
CGGCAAGACCAAAGTCCACAGCCCGGTCTGCCCGTTCTACGGAAACCCTAGCCCGCCTGCCC
CATGTCCCTGCCCGCTCCGACAGGCTTGGGGGAGCCTCGACGCGCTCGTCGGAAGGCTAAG
GGCAGCGTTCGAAGAGAATGGAGGAAAACCAGAGATGAACCCTTTCGGTGCAAGAGCCGTC
AGGCTCTATTTACGAGAAGTTCGTGACGTGCAGAGCAAGGCTCGAGGCGTCAGCTACGAGA
AGAAGAAGAGGAAGCGCCCGGCGGCAGCGGCTCAAGGCGGCGCGGCGGGTGACGAGGG
CAGCGATCAGCAGCCGGATACGTCTCCAAGTGAAGGGTAG

B

Yellow target 1 Yellow target 2

WT: ——ACCTCCACCACCACCAC-CGCCGG——GACGTGCAGAGCAAGG-CTCGAGG——
3#: ——ACCTCCACCACCACCACACGCCGG——GACGTGCAGAGCAAGGACTCGAGG——
19#: ——ACCTCCACCACCACCACACGCCGG——GACGTGCAGAGCAAGG-CTCGAGG——

Gray target 1 Gray target 2

WT: ——CCCATCA-TTGTTCCCCAATCCTC——CCAACACCTACTCTGCAGCGACA——
40#: ——CCCATCAATTGTTCCCCAATCCTC——CCAACACCTACTCTGCAGCGACG——

Gray target 3

WT: ——CAAGACCAAAGTCCACA-GCCCGG——
40#: ——CAAGACCAAAGTCCACAAGCCCGG——

Fig. S5. CRISPR/Cas9 genome editing. (A) The genomic DNA sequence of *TfALOG3*. The black and red characters represent exons and introns, respectively; PAMs (Protospacer Adjacent Motifs) are highlighted in green. Two CRISPR/Cas9 plasmids were used in this study, and the different CRISPR/Cas9 targets are highlighted in yellow and gray, respectively. Start and stop codons are highlighted in red. (B) Mutations in the WT and three *TfALOG3*-Cas9 plants.

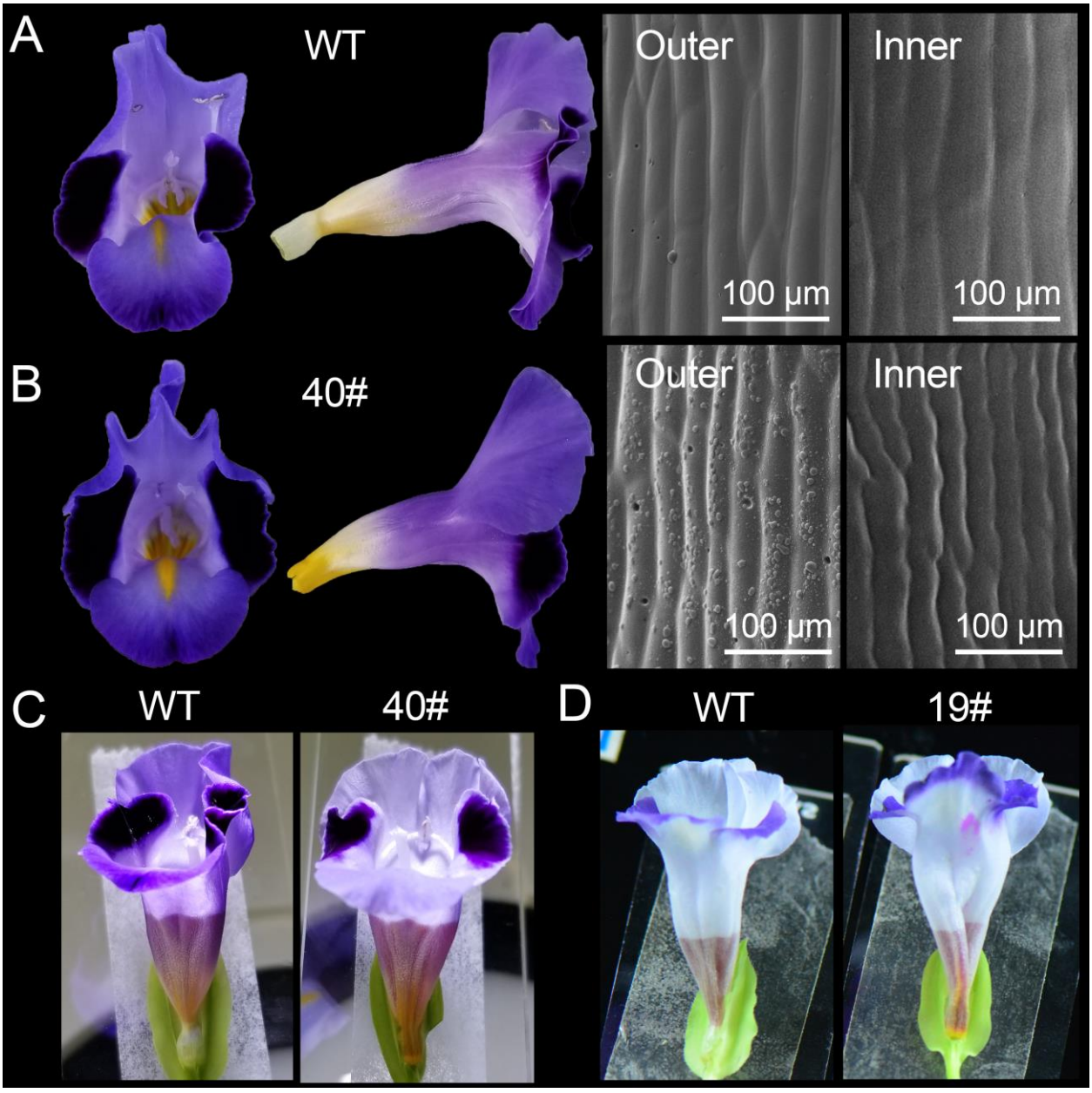

Fig. S6. Function and epidermal morphology of the corolla neck in the 19# and 40# transgenic lines. (A, B) Front and side views of *Torenia fournieri* wild-type (A) and *TfALOG3*-Cas9 40# (B) flowers. The outer and inner epidermal cell morphologies are also shown. (C, D) Application of 200 μl water containing 0.1% Safranin O to different flowers.

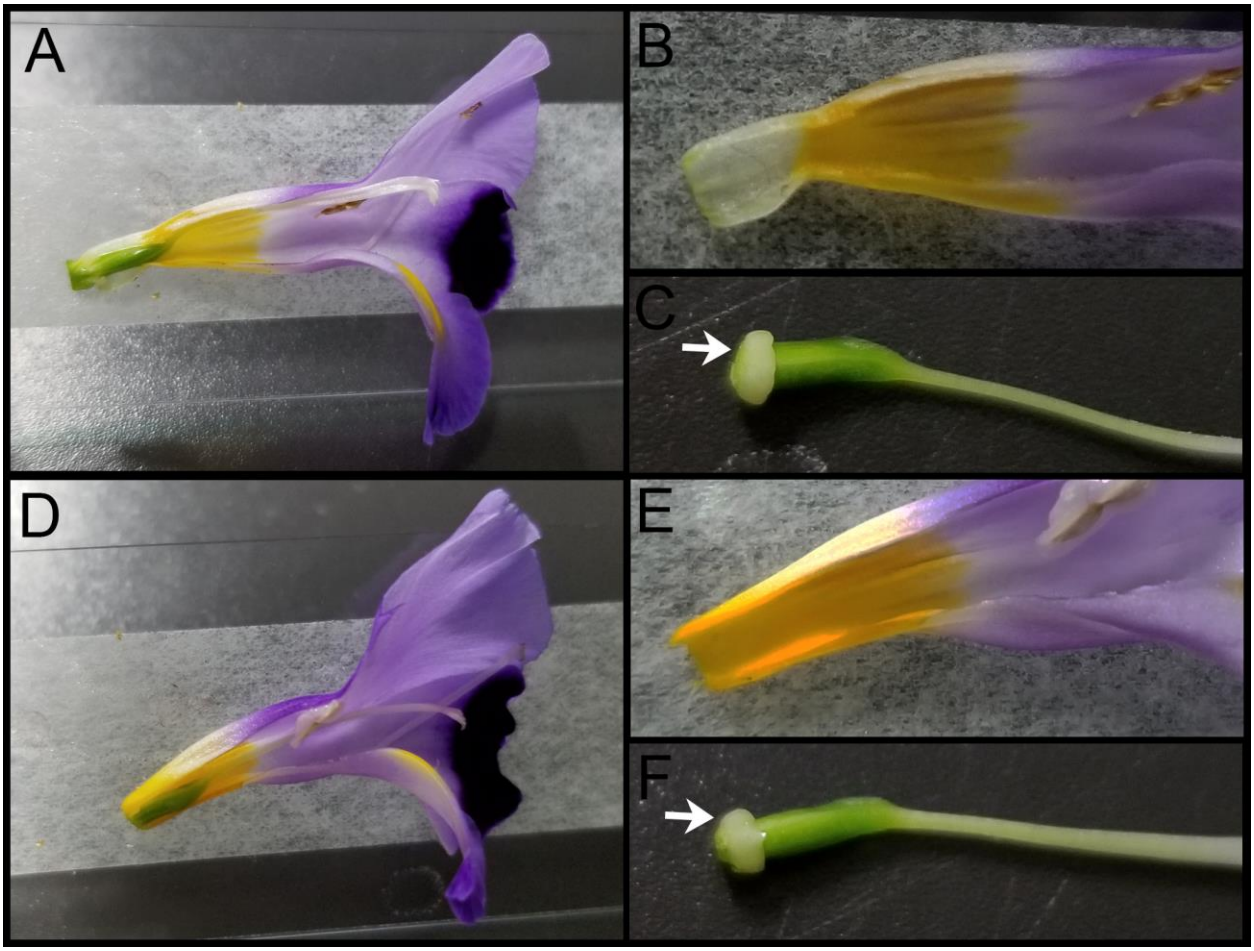

Fig. S7. Longitudinally dissected flowers from the WT and mutant plants. (A–F) Dissected WT (A–C) and 40# mutant (D–F) flowers. White arrows indicate the nectary in the floral base surrounding the ovary.

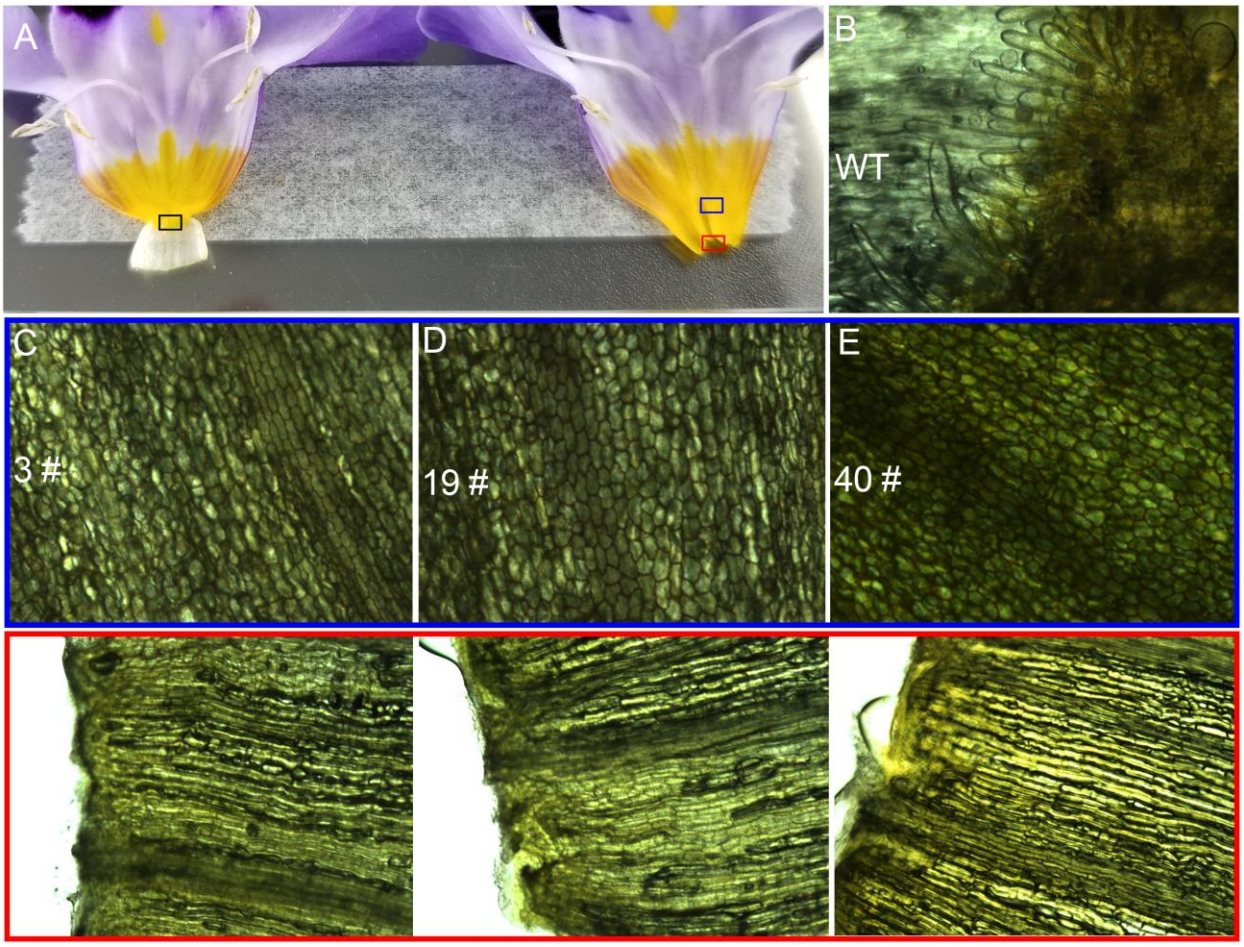

Fig. S8. Inner epidermis of the WT and mutant corolla bottoms. (A) Open corolla tubes from the WT (left) and mutant (right) plants. Black box indicates the regions being imaged in (B); blue and red boxes indicates the regions being imaged in (C-E). (B–E) Images of the WT corolla neck (B) and mutant corolla bottom (C–E).

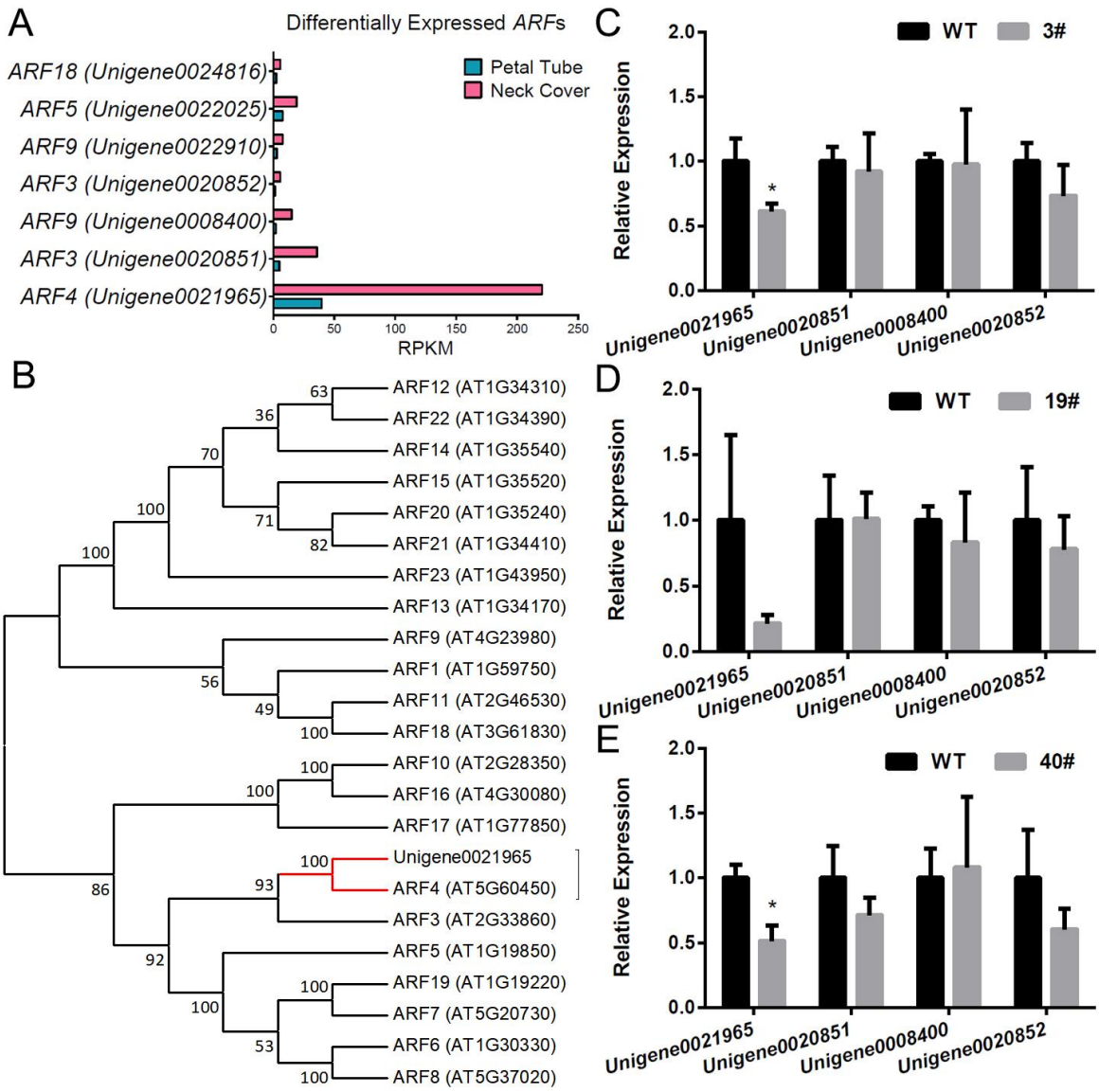

Fig. S9. Expression of *AUXIN RESPONSE FACTOR* (*ARF*) genes and a phylogenetic tree for ARF4. (A) Seven *AUXIN RESPONSE FACTOR*s enriched in the corolla neck according to transcriptomic analyses. The reads per kilobase per million mapped reads value for each unigene are shown. (B) A neighbor-joining tree for ARF4 together with all Arabidopsis ARF proteins. The red branch shows the relationship between Unigene0021965 and ARF4. The bootstrap value (percentage, based on 1000) is marked at each node, and the accession numbers are indicated as the suffix of each sequence. (C–E). Real-time PCR analysis of four *ARF* genes enriched in the corolla neck of the WT and three transgenic lines. The error bars are ± 1 SD from three biological replicates; asterisks indicate significant differences between the mutant and WT plants ($P < 0.05$).

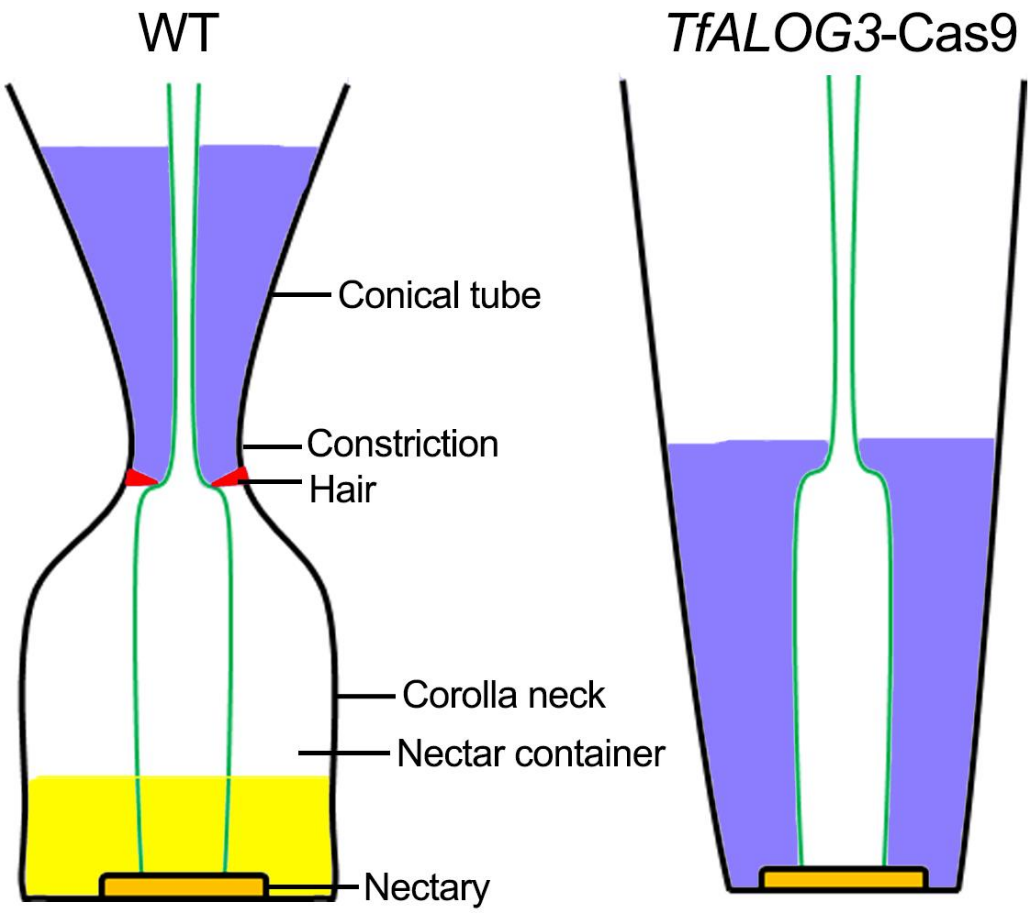

Fig. S10. Model of how the corolla neck prevents water from reaching the nectar container. The corolla, pistil, and nectary are shown in both the WT and mutant plants. Hairs in the constriction at the top of the corolla neck in WT plants are shown as red triangles; the blue block represents external water and yellow block the undiluted nectar.

Table S1. Primers used in this study.

Click here to Download Table S1

Table S2. Reads numbers, RPKM, FDR and annotation of differentially expressed genes.

Click here to Download Table S2