

Supplementary Materials

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Figure S5. Comparison of flower tube size in tobacco

Table S1. Sequences of primers used for gene cloning and genomic DNA PCR

Gene	Encoded protein	Primer sequence (5'-3')		GenBank
<i>MdTFL1</i>	TFL1-like protein	F	GGGGTACCATGAAAAGAGCCTCGGAG	AB052994.1
		R	CGGGATCCCTAGCGTCTTCTAGCTG	
<i>hptII</i> <i>563bp</i>	hygromycin phosphotransferase gene	F	CTCGGAGGGCGAAGAATCTC	KU561939.1
		R	CAATGACCGCTGTTATGCGG	

Table S2. Sequences of primers used for qRT-PCR in *Nicotiana tabacum*

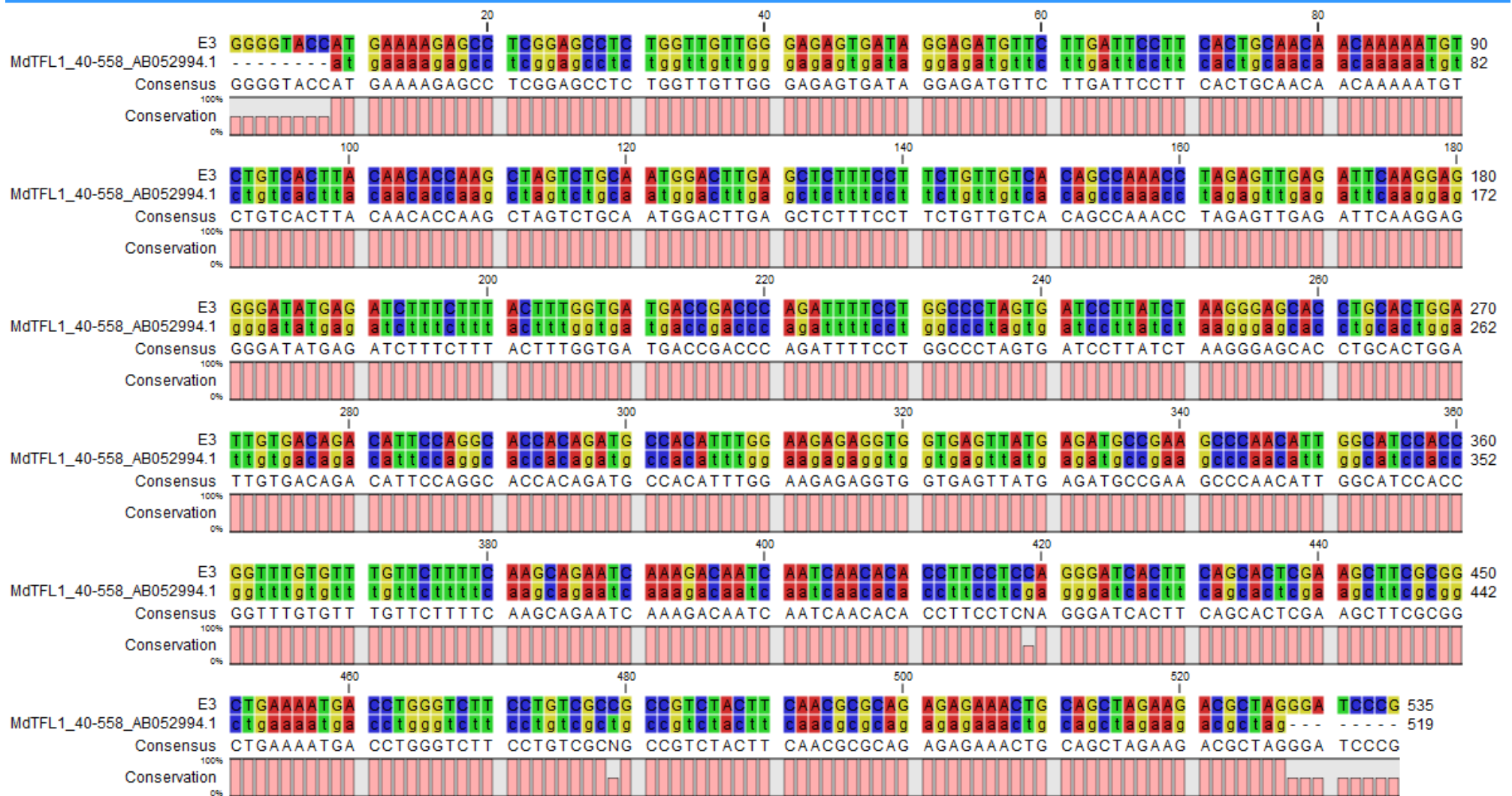
Gene	Encoded protein		Primer sequence (5'-3')	GenBank
<i>NtAct</i>	Actin (Tob103)	F	TGGCTCAACCATGTTCCCTG	U60495.1
		R	CCGGTGGAGCAACAACCTTA	
<i>NtAPI</i>	Apetala 1 protein	F	TCTCAGTGCTTTGTGACGCT	JQ686939.1
		R	GCGTCTCTCTGCGTATGAGT	
<i>NtSOC1</i>	MADS-box protein SOC1-like	F	GGTGGCCTTCAACAGAGACA	JQ686938.1
		R	GGGACGCCTTATTCTGCACT	
<i>NFL1</i>	LEAFY/FLORICAULA (LFY)	F	GCTTCTAGCTCGGTTTCTGGT	JQ686928.1
		R	AGGGGCAAGCAAAGTCAGAA	
<i>NtFT1</i>	Flowering locus T1	F	CAACCAACCTAGGGTTGACATT	JX679067.1
		R	GTTTGGGTTGCTTGGGGTTG	
<i>NtFT2</i>	Flowering locus T2	F	TTCGGCAATTGGATCGAGAGG	JX679068
		R	GGCAACAGGCAAATTGAGACC	
<i>NtFT3</i>	Flowering locus T3	F	GGAGCACACTTCGGAATGA	JX679069
		R	TCGAGTCAATTGTGAAACAGC	
<i>NtFT4</i>	Flowering locus T4	F	GGCGTCAGAATTTGAGCACA	JX679070
		R	GCAATTGAAGTAAACAGCAGAAACG	
<i>NtCET2</i>	CEN-like protein 2	F	ACGCGTAAATTCGCAGAAGA	AF145260.1
		R	TTCTGGCAGCAGTTTCCCTC	
<i>NtCET4</i>	CEN-like protein 4	F	GGTATGAAATGCCAAGGCCA	AF145261.1
		R	TCTTCTGAGAATTTGCGCGT	
<i>NtCCD8</i>	Carotenoid cleavage dioxygenase 8	F	AGTTGTAGAGGAGGACCAGCA	NM_001325897.1
		R	CTTAGTGGCATTTCGGGCAC	
<i>NtCCD1-3</i>	Carotenoid cleavage dioxygenase 1-3	F	CTCGATTGAGTCCCTCCCG	KM605431.1
		R	ATCTCCCTCCTCCCAAGCAT	
<i>NtCCD4-1</i>	Carotenoid cleavage dioxygenase 4-1	F	GGATCCCAACAATTCGGCAG	KM605432.1
		R	GGGGACTTTGCATCCATGAC	
<i>NtCCD4-2</i>	Carotenoid cleavage dioxygenase 4-2	F	CAGACTCGGGGAAAATCCCT	KM605433.1
		R	ATTGATCCCGGGCACATCAA	

Table S3. Leaf Mass Per Area (LMA) measurements

Genotype	Leaf No	Area	Subtotal	Max Width (cm)	Average Width (cm)	Length (cm)	Length/Max Width Ratio
WT	1	90.2	90.2	8.1	4.6	19.5	2.41
	2	115.2	205.4	9.6	5.4	21.5	2.24
	3	97.4	302.7	8.8	5.5	17.8	2.02
	4	133.4	436.1	9.8	6.2	21.5	2.19
	5	68.5	504.6	7.3	4.4	15.4	2.11
	6	65.4	570.0	6.8	4.2	15.7	2.31
	7	94.1	664.1	8.2	5.2	18.2	2.22
Average		94.9	396.2	8.4	5.1	18.5	2.21
<i>35S:: MdTFL1</i>	1	52.3	52.3	5.9	3.4	15.4	2.61
	2	53.0	105.3	5.6	3.3	16.2	2.89
	3	69.4	174.7	6.5	3.9	17.9	2.75
	4	59.8	234.5	5.9	4.0	15.0	2.54
	5	89.3	323.7	7.3	4.5	19.8	2.71
	6	71.4	395.1	6.4	4.2	17.0	2.66
	7	49.1	444.1	5.9	3.4	14.5	2.46
	8	68.4	512.6	6.4	4.0	17.2	2.69
	9	38.6	551.2	4.5	2.4	16.3	3.62
	10	34.6	585.8	4.6	2.7	13.0	2.83
Average		58.6	337.9	5.9	3.6	16.2	2.78

CDS: 40..558

Figure S1. DNA sequence of *MdTFL1* cloned in this study. CDS region starts with **ATG** and stops at **TAG**, underlined sequences were used for design-specific primers.



Alignment of isolated *MdTFL1* (E3) to the known sequence of original DNA (GenBank: AB052994.1)

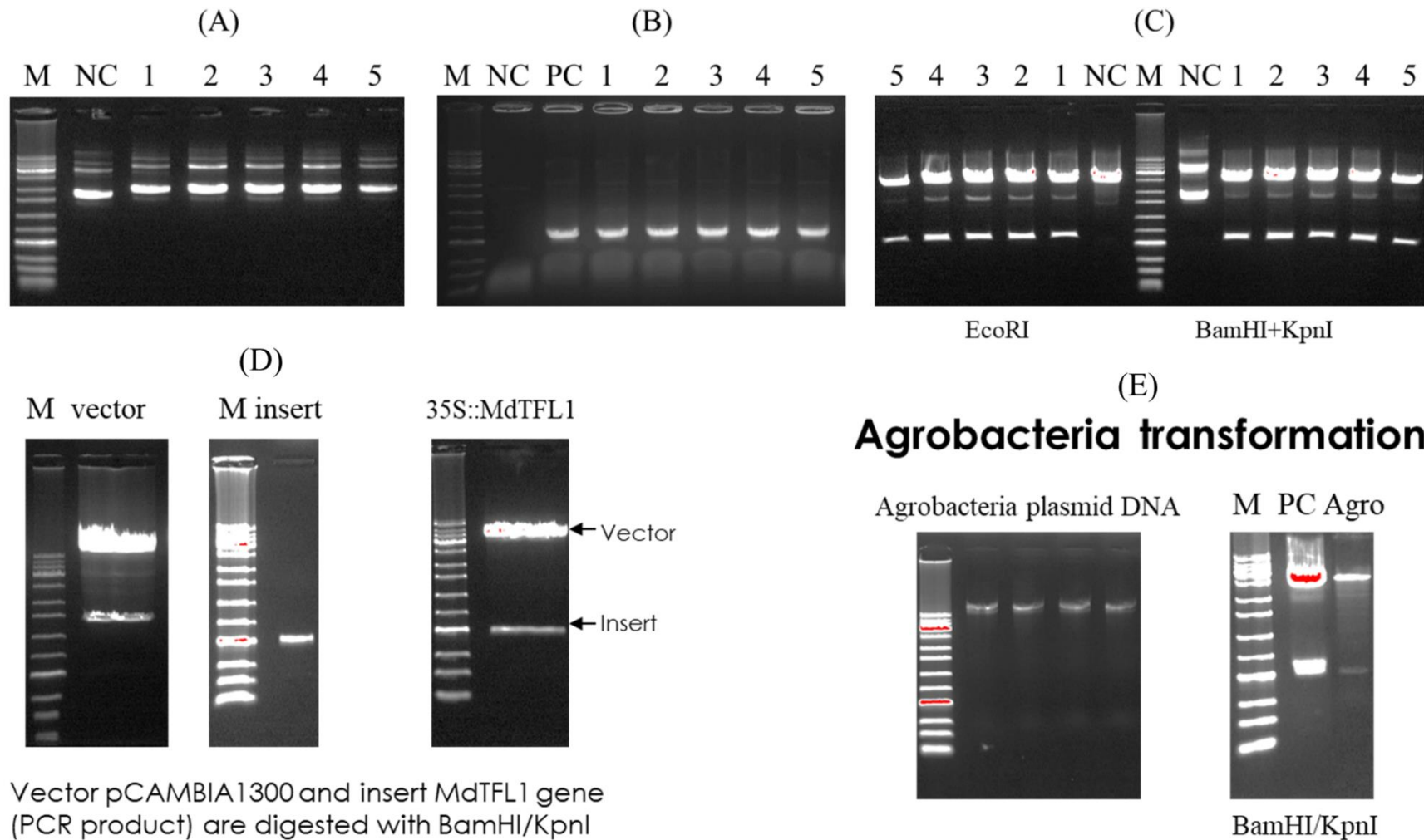


Figure S2. Gene cloning and expression vector construction. Isolated CDS of MdTFL1 containing a KpnI site at the 5' end and a BamHI site at the 3' end were used for gene cloning. (A) Plasmid DNA of *MdTFL1* in pGEM Teasy vector. (B) Colony PCR of transformed *E. coli* harboring MdTFL1::pGEM Teasy. (C) MdTFL1::pGEM Teasy was digested with EcoRI and BamHI+KpnI. (D) *MdTFL1* was ligated to binary vector pCambia1300 resulting in 35S::MdTFL1 expression vector and confirmed by BamHI+KpnI double enzyme digestion. (E) Transformation of the expression vectors into *A. tumefaciens* EHA105. The plasmid DNA of expression vector 35S::MdTFL1 in the transformed *A. tumefaciens* EHA105 was isolated and then double-checked by restriction enzyme digestion BamHI+KpnI.

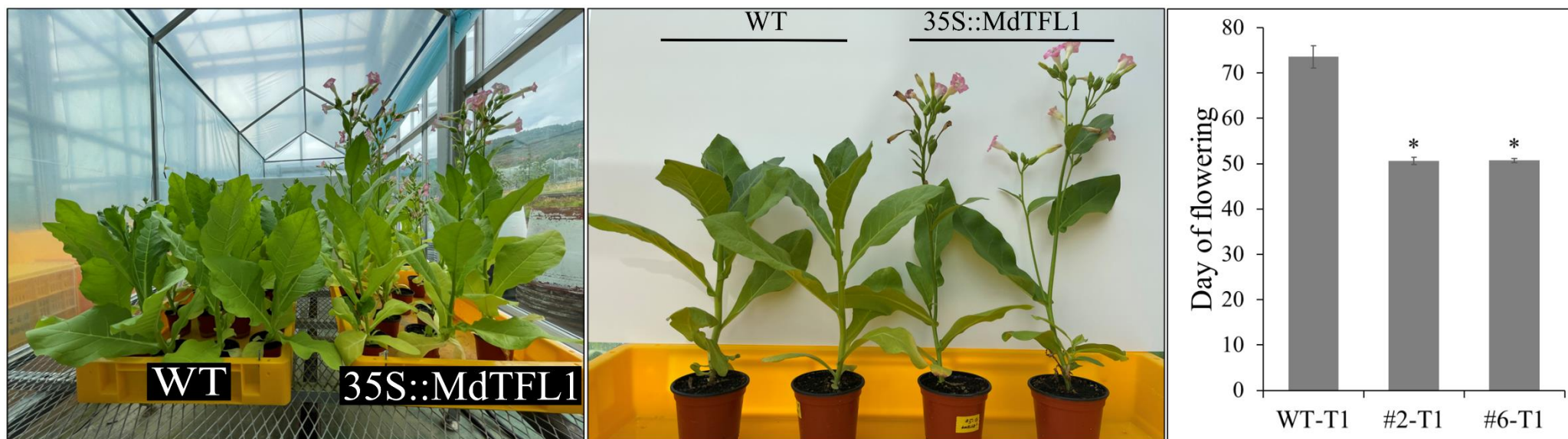


Figure S3. Phenotype and flowering time in T1 plants

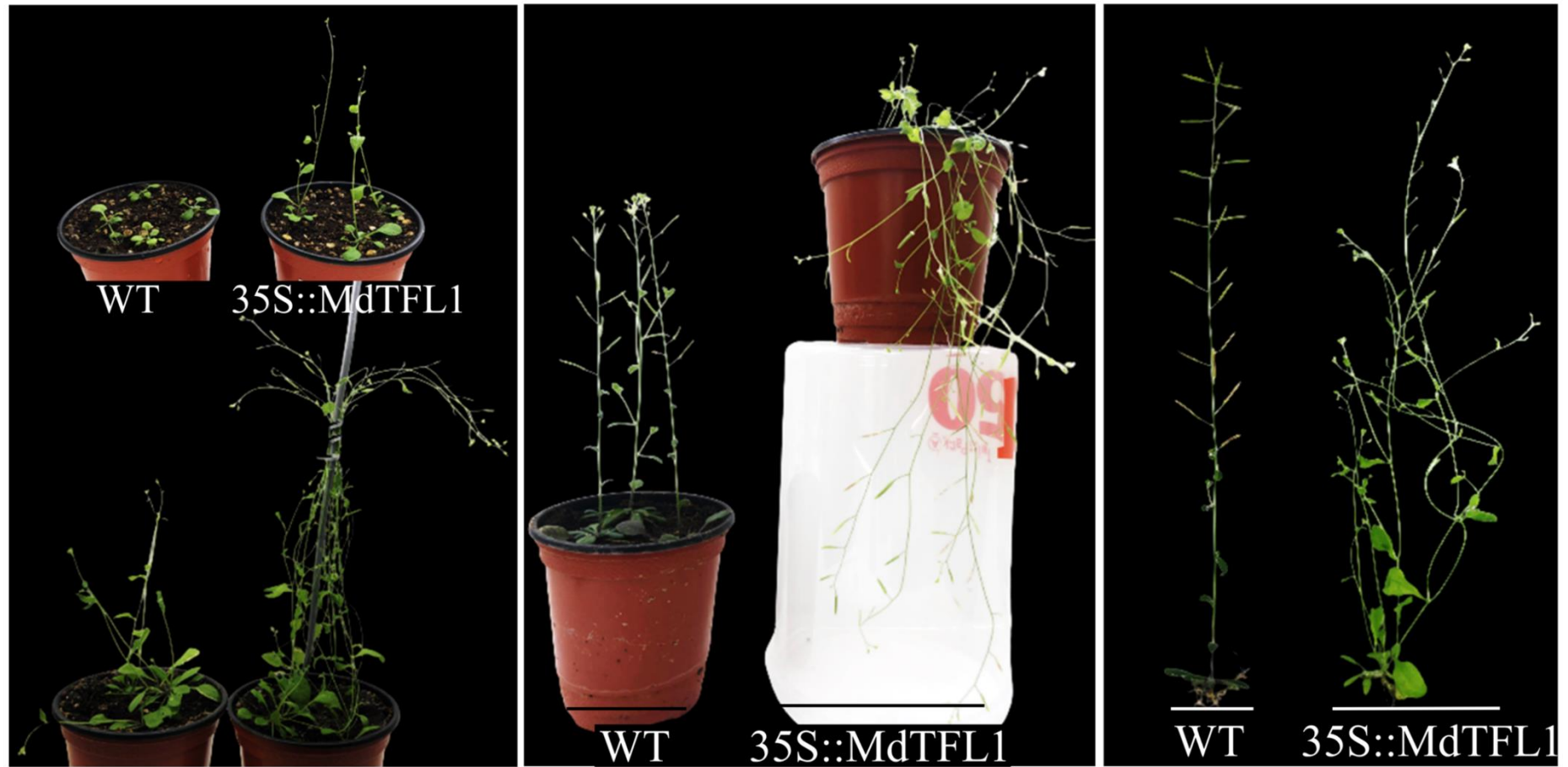
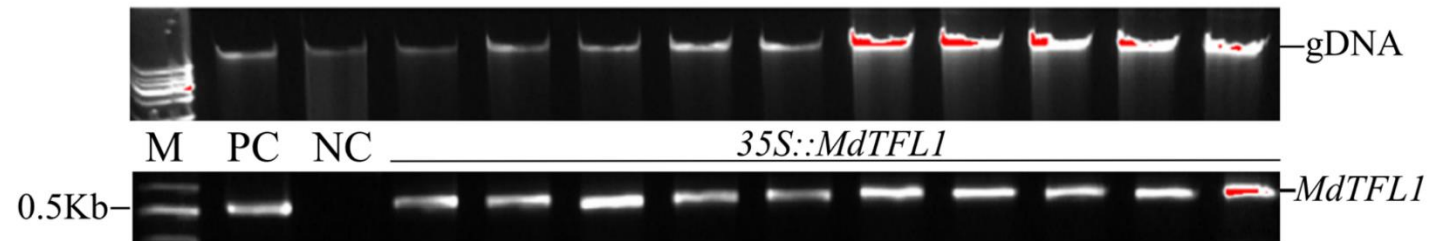


Figure S4. Antisense expression of MdTFL1 in *Arabidopsis thaliana*

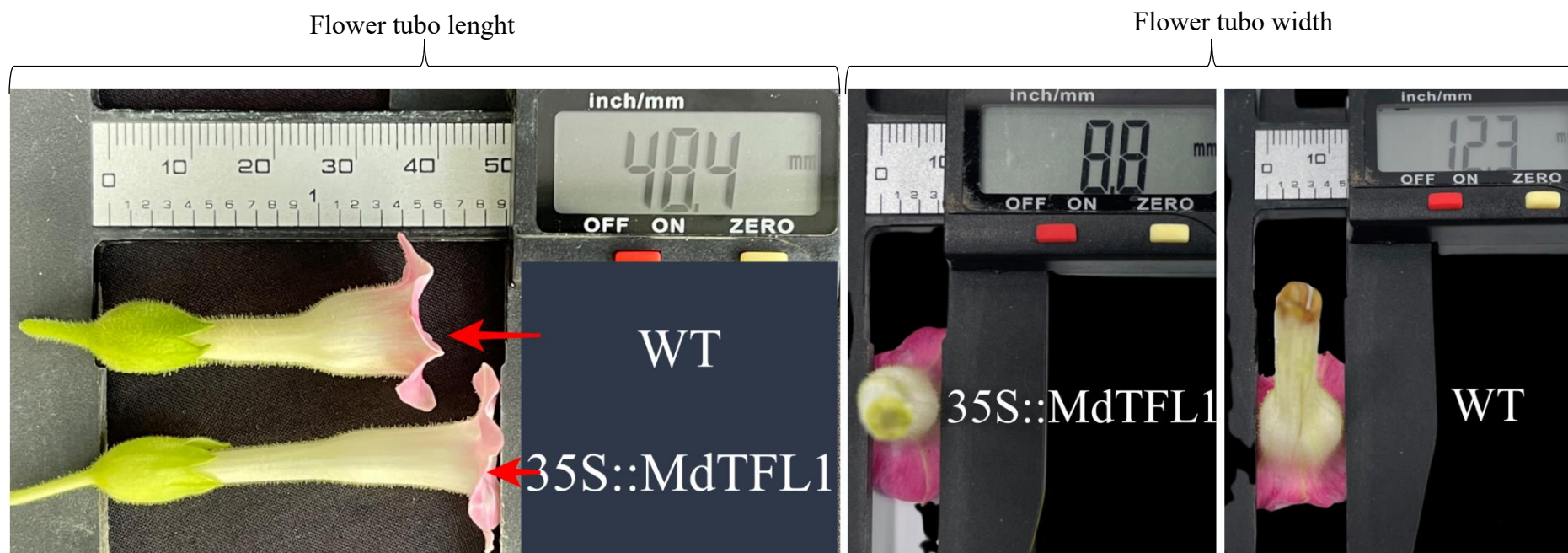


Figure S5. Comparison of flower tubo size in tobacco