

Supplementary Material

PaNAC089 is a membrane-tethered transcription factor (MTTF) that modulates flowering, chlorophyll breakdown and trichome initiation

Changsheng Shao^A, Fangfang Cai^{A,B}, Zhiru Bao^A, Yanping Zhang^A, Gehui Shi^A, Zheng Zhou^A, Xiyang Chen^A, Yangyang Li^A, Manzhu Bao^{A,}, and Jiaqi Zhang^{A,*}*

^AKey Laboratory of Horticultural Plant Biology, Ministry of Education, College of Horticulture and Forestry Sciences, Huazhong Agricultural University, Wuhan 430070, Hubei, China.

^BPlant Genomics & Molecular Improvement of Colored Fiber Laboratory, College of Life Sciences and Medicine, Zhejiang Sci-Tech University, Hangzhou 310018, Zhejiang, China.

*Correspondence to: Manzhu Bao Key Laboratory of Horticultural Plant Biology, Ministry of Education, College of Horticulture and Forestry Sciences, Huazhong Agricultural University, Wuhan 430070, Hubei, China Email: mzbao@mail.hzau.edu.cn and Jiaqi Zhang Key Laboratory of Horticultural Plant Biology, Ministry of Education, College of Horticulture and Forestry Sciences, Huazhong Agricultural University, Wuhan 430070, Hubei, China Email: jiaqizhang@mail.hzau.edu.cn

Supplementary Material

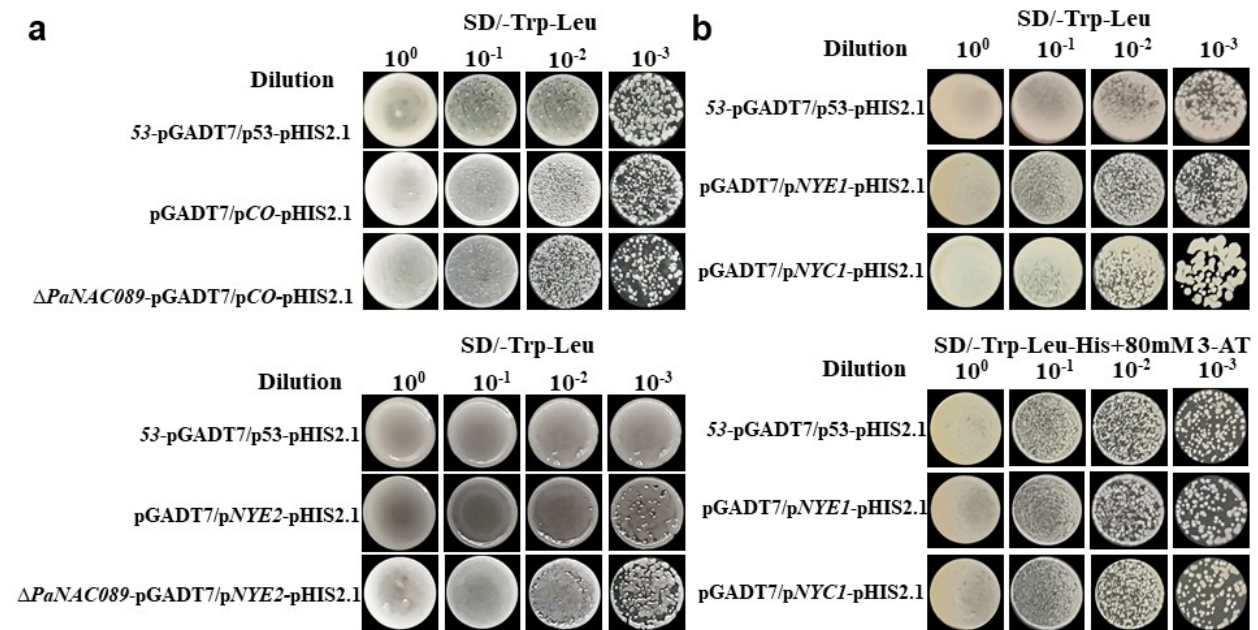


Fig. S1. The result of yeast one-hybrid assays. Different concentrations (cell/mL) were cultured on SD/-Leu-Trp medium. **a** The double dropout controls of yeast one-hybrid assays of Δ PaNAC089 with the promoters of *CO* and *NYE2*. **b** Yeast one-hybrid assays of Δ PaNAC089 with the promoters of *NYC1* and *NYE1*. Repeated thrice.

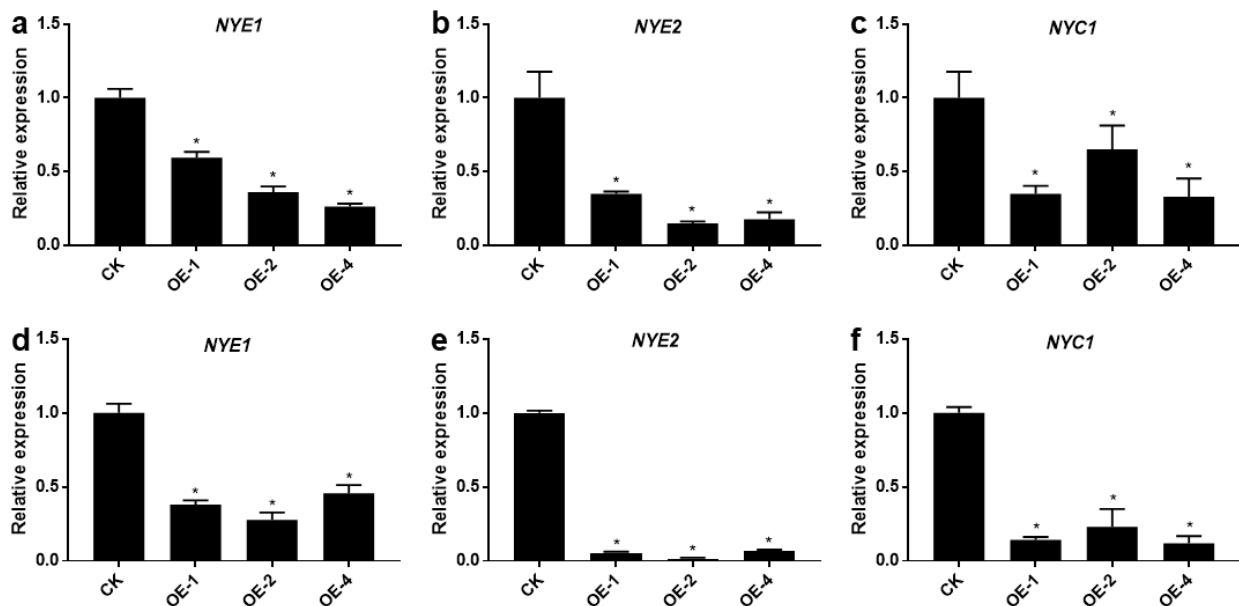


Fig. S2. $\Delta PaNAC089$ inhibits CCGs expression in Arabidopsis. **a-c** Expression analysis of some CCGs (*NYE1*, *NYE2*, *NYC1*) in 20-day-old CK and 20-day-old transgenic lines ((OE-1, OE-2, OE-4)). Data are mean \pm SD. **d-f** Expression analysis of some CCGs (*NYE1*, *NYE2*, *NYC1*) in 40-day-old CK and 40-day-old transgenic lines (OE-1, OE-2, OE-4). Data are mean \pm SD. For (**a-f**) error bars are standard deviation (SD) and * indicates $P \leq 0.05$. Each process was repeated thrice.

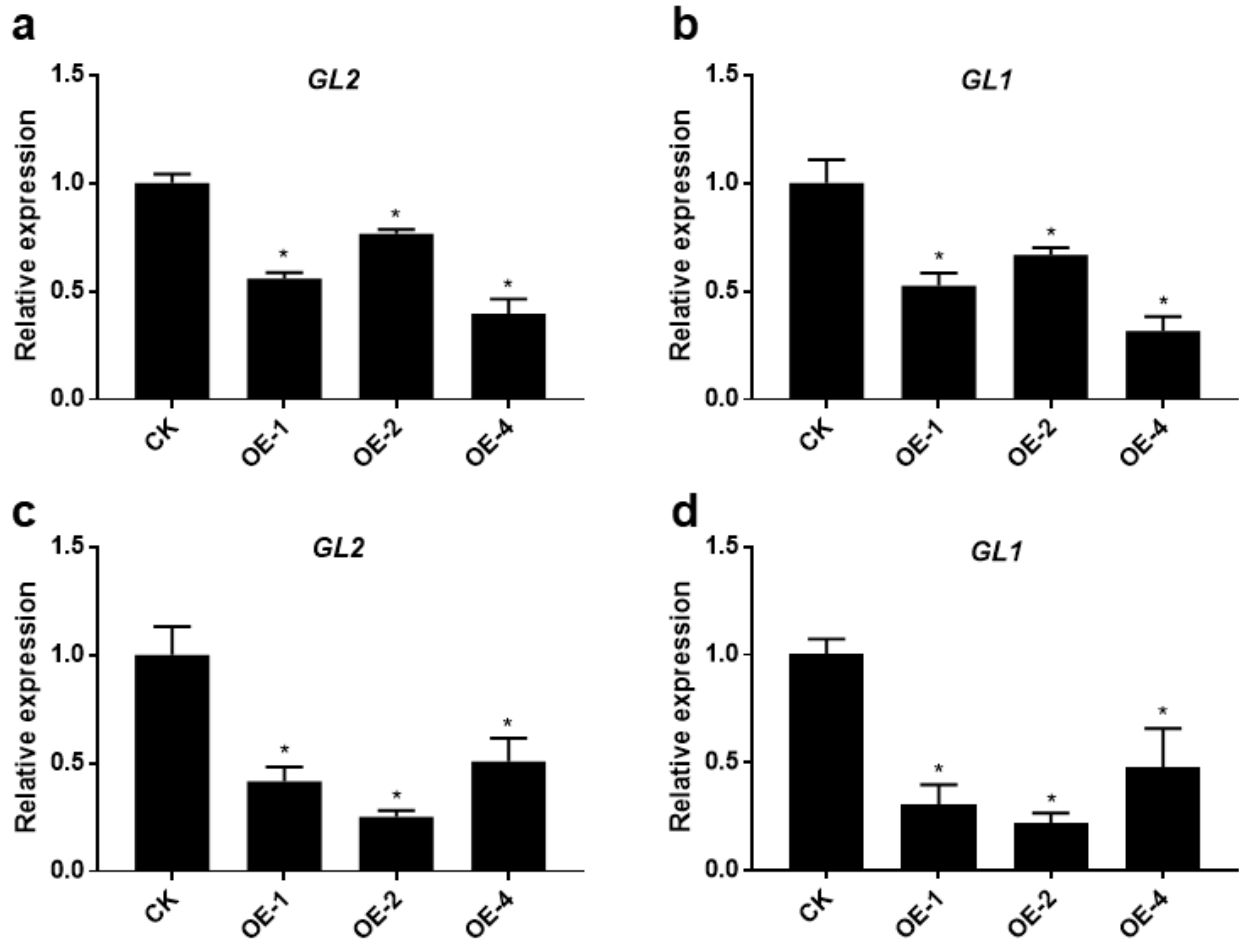


Fig. S3. $\Delta PaNAC089$ inhibits trichomes related genes expression in Arabidopsis. **a, b** Expression analysis of some trichomes related genes in CK and transgenic lines of 15-days-age (OE-1, OE-2, OE-4). Data are mean \pm SD. **c, d** Expression analysis of some trichomes related genes in CK and transgenic lines of 20-day-age (OE-1, OE-2, OE-4). Data are mean \pm SD. For (**a-d**) error bars are standard deviation (SD) and * indicates $P \leq 0.05$. Each process was repeated thrice.

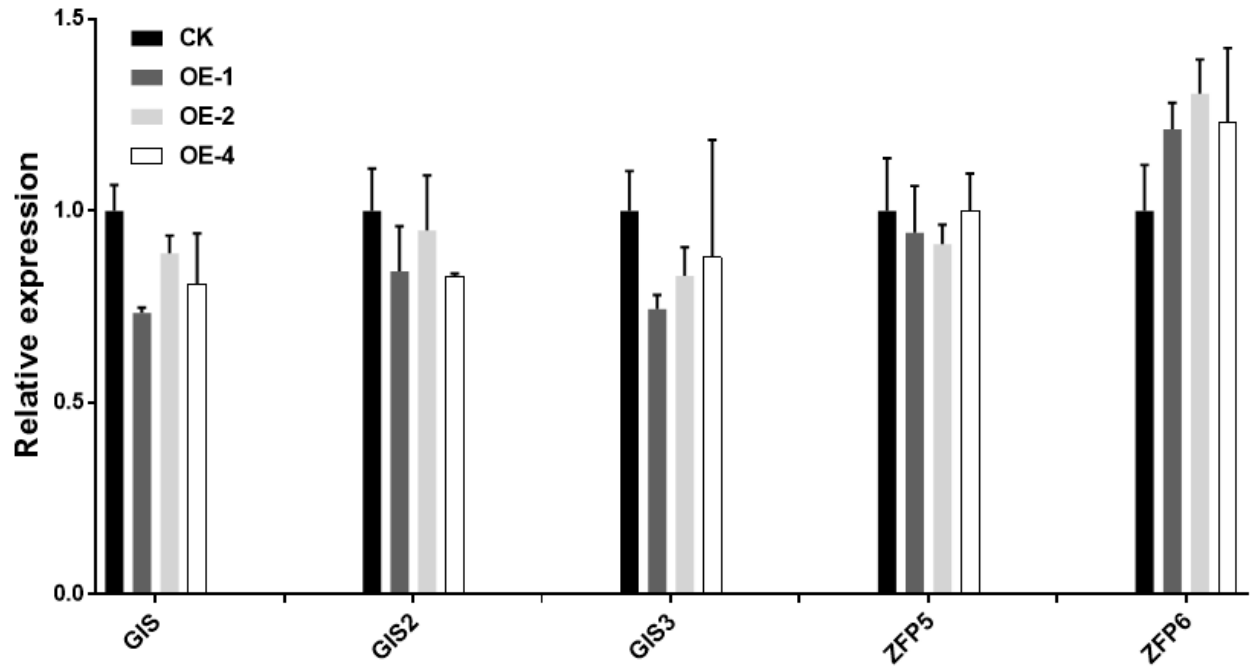


Fig. S4. Expression analysis of *GIS* family genes in CK and transgenic lines (OE-1, OE-2, OE-4) of 10-day-age. Data are mean \pm SD.

Table S1. The primers used in this study

pCAMBIA2300s	size of amplicons	Primer sequence
PaNAC089-F	1182bp	GGTACCATGGCGGACACTTCTCGGTTCCCCG
PaNAC089-R		GTCGACTTACGACATGATATTAGATATAAAC
Δ PaNAC089-F	1077bp	GGTACCATGGCGGACACTTCTCGGTTCT
Δ PaNAC089-R		GTCGACTTAGTTCTTGACTGTCCAAACTG
Yeast one-hybrid assays		Primer sequence
Δ PaNAC089-AD-F	1077bp	GAATTCATGGCGGACACTTCTCGGTTCCCCG
Δ PaNAC089-AD-R		GGATCCTTTACGACATGATATTAGATATAAAC
pAtNYE1- pHIS2.1F	117bp	AATTCATGACGACACATGGGGAAGCTCATGCAAAGATTAGAGAACACGTGGCACTCTCTCGTGTTCAGAAAATTCCAAAAGAGTGTTCAGATTGGGCACGGAAGTGTGGAGCTCCACACTTCGGTGCCCAAATCTGAACACTCTTTTGGAAATTTTCTGAACACGAGAGAGTGCCACGTGTTCTCTAATCTTTGCATGAGCTTCCCCATGTGTCGTCATGG
pAtNYE1- pHIS2.1R		
pAtNYE2- pHIS2.1F	93bp	AATTCAAAGCCACGTGTGAACCTTAATCTCAGCACACATGCTAAGCTCATGCAAGTGAAGAAGCTGAACACCACGTCCTCGTGGCACTCGAGCT
pAtNYE2- pHIS2.1R		CGAGTGCCACGAGGACGTGGTGTTCAGCTTCTTCACTGCATGAGCTTAGCATGTGTCGTGAGATTAAGGTTCCACACGTGGCTTTG
pAtNYC1- pHIS2.1F	321bp	GAATTCAAAATTTTGTTCATGTGAACC
pAtNYC1- pHIS2.1R		GAGCTCGGCCACACGTTACAGCAACC
Subcellular localization		Primer sequence
PaNAC089-YFP-F	1182bp	GGATCCATGGCGGACACTTCTCGGTTCT
PaNAC089-YFP-R		GGTACCCGACATGATATTAGATATAAA
Δ PaNAC089-YFP-F	1077bp	GGATCCATGGCGGACACTTCTCGGTTCT

ΔPaNAC089-YFP-R		GGTACCCAGAAACAAAACCAGCAAAGC
Quantitative real-time PCR		Primer sequence
PaNAC089-QTF	83bp	GGGAGAGGACAGATTGGATAA
PaNAC089-QTR		GTCCGAAGGCGACAAACAACA
AtNYE1-QTF	101bp	GCAAGGATGGGCAAATAGG
AtNYE1-QTR		CACCGCTTATGTGACAATGAAC
AtNYE2-QTF	97bp	GACGAAGTAGTGGCGGAGTG
AtNYE2-QTR		CGATGAGATTCAAGAAGAAGTGG
AtNYC1-QTF	130bp	TTCTCAGTGGTTTCGAGCATT
AtNYC1-QTR		AGGTAATTGACGGCTTTTCC
AtCO-QTF	127bp	GCCATCAGCGAGTTCCAATTCTAC
AtCO-QTR		CCTTCCTCTTGATCCACCACCAG
AtFT-QTF	114bp	TCCCTGCTACAACCTGGAACAACCT
AtFT-QTR		GCCTGCCAAGCTGTGAAACAATA
AtSOC1-QTF	153bp	TAAGGATCGAGTCAGCACCAAACC
AtSOC1-QTR		AGCTCCTCGATTGAGCATGTTCT
AtAP1-QTF	110bp	AAATCCAGCATCCTTACATGCTCTC
AtAP1-QTR		CAGTTCGAGATCATTCTCCTCATT
AtLFY-QTF	131bp	TACTCTCCGCGCTGGTGATTG
AtLFY-QTR		ACTTCCTCCTCCGCCGTTATTCC
Luciferase reporter system assay		Primer sequence
ΔPaNAC089-F	1077bp	GGTACCATGGCGGACACTTCTCGGTTC
ΔPaNAC089-R		GTCGACTTAGTTCTTGACTGTCCAAACTG
pAtNYE1-LUC-F	1592bp	ctatagggcgaattgggtaccCCACAAGAAACACCAATAGCAAAC
pAtNYE1-LUC-R		aagcttatcgataccgtcgacCTCTGCTCTCTTGAAACCCAAATC
pAtNYE2-LUC-F	1518bp	ctatagggcgaattgggtaccATTCCAATCCCATATATCACGCA
pAtNYE2-LUC-R		aagcttatcgataccgtcgacCTTTGCTTGTTCTCAAAAATATCCG
pAtNYC1-LUC-F	1413bp	ctatagggcgaattgggtaccGCCATTCCAACCTCATTTAGTAT
pAtNYC1-LUC-R		aagcttatcgataccgtcgacTAGAAAACAAGATAAACGAAGGAGCT