

Supplemental Data

Analysis of Arabidopsis *glucose insensitive growth* Mutants Reveals Involvement of the Plastidial Copper Transporter PAA1 in Glucose-induced Intracellular Signaling

Lee et al.

Supplemental Figure S1. Analysis of the stem cell niche in Col-0 and *gig* roots. Cell type-specific markers were used to monitor defects in and around the stem cell niche. A, *pSHR::SHR-GFP*. B, *pSCR::GFP-SCR*. C, *QC25::GUS*. D, *pWOX5::GFP*.

Supplemental Figure S2. Comparative analysis of Col-0 and *gig* adult plants. A, Top view. B, Side view. Rosette leaves of *gig* are smaller and inflorescence stems of *gig* are shorter than those of Col-0. F₁ progeny of crosses between Col-0 and *gig* (Col-0 x *gig*) are indistinguishably similar to Col-0.

Supplemental Figure S3. Complementation of *gig* with Cu supplementation. A, DIC image of root meristem size of Col-0 and *gig* at 8 DAG in the presence of 10 μ M CuSO₄. Black arrowheads indicate the QC, whereas blue arrowheads demarcate the upper border of the MZ. Measurements of meristem cortex cell number (B) and meristem length (C) from 2 to 10 DAG in the presence of 10 μ M CuSO₄.

Supplemental Figure S4. Root growth assay in the absence or presence of Cu with increasing Glc concentrations. Twelve-day-old seedlings of Col-0 (blue) and *gig* (red) were grown on MS agar plates without (A) and with 10 μ M CuSO₄ (B). Error bars indicate the SE of mean from biological triplicates.

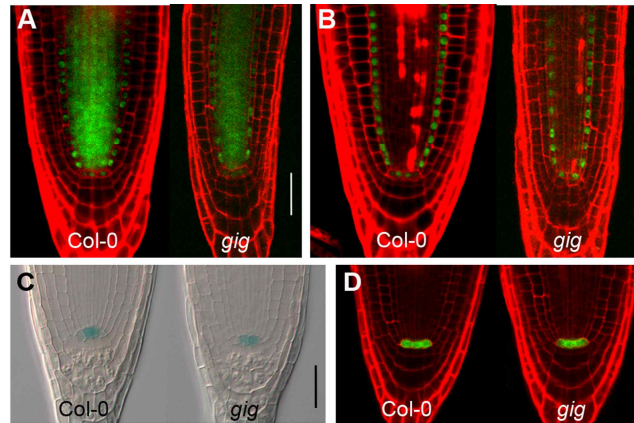
Supplemental Figure S5. Expression analysis of Glc-responsive genes with the addition of Cu. qRT-PCR in Col-0 and *gig* in the presence of 1% (blue) and 6% (red) Glc with 10 μ M CuSO₄. A, *APL3*. B, *HXK1*. C, *ABI4*. Statistical significance of differences was determined by Student's *t*-test (asterisk for *P*<0.05). Error bars indicate the SE of mean from biological triplicates.

Supplemental Figure S6. Prediction of an ABRE sequence in the *GIG* promoter. The combination of a CCAC/ACGT core element that is known for both ABI4 binding and retrograde signaling is underlined.

Supplemental Table S1. Sequence information of PCR primers used in this study

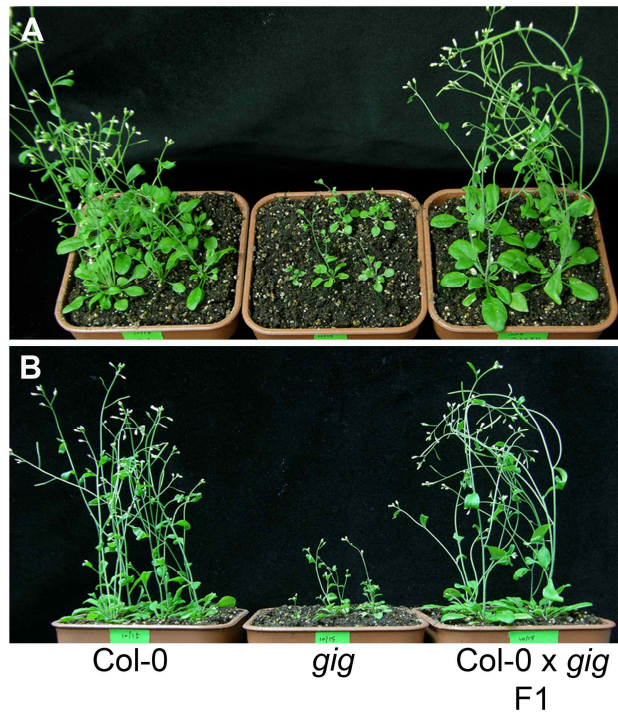
Purpose	Name		Sequence (5'–3')
TAIL-PCR	AD2		NGTCGASWGANAWGAA
	AD5		SSTGGSTANATWATWCT
	LB50		ATAATAACGCTGCGGACATCT
	LB100		CCTATAAATACGACGGATCGT
	LB150		CACGTCGAAATAAAGATTTCCGA
Genotyping	gig-1	LP	ACCTTGTCAACCAACTGCTG
		RP	GGTATGTAGTAGCTGCGTCT
	pSKI015	RB1	CTAGATCCGAAACTATCAGTG
	gig-2	LP	AACCCTCTCAAGACCAAGAGC
		RP	ATTGGGCTGTAGGCATAAACC
	SALK_070739	LP	TTGTTTTTGATTCCAAATCGG
RP		TCATCAAATGAGGAGGAATCG	
SALK	LB	GGCAATCAGCTGTTGCCCGTCTCACTGGTG	
abi4-1	F	GGACCCCTTAGCTTCCCAAC	
	R	CGGATCCAGACCCATAGAAC	
pGIG::GUS	pGIG	F	CACCAGAACAAGAAAATACTTATCCGAGC
		R	GAAAGTGGGTGGCTCCGTA
pGIG::GIG-GFP	pGIG GIG	F	CACCAGAACAAGAAAATACTTATCCGAGC
		R	AGAGCTTTGCTTCCATCTTG TG
Transient expression assay	pGIG	F	AAGCTTAGAACAAGAAAATACTTATCCG
		R	TCTAGAGAAAGTGGGTGGCTCCGTA
ABI4	ABI4	F	TCTAGAATGGACCCCTTAGCTTCCCAAC
		R	GAGCTCTTAATAGAATCCCCCAAGATGG
qRT-PCR	ACT2	F	TCGCTGACCGTATGAGCAAAGAA
		R	TGGAATGTGCTGAGGGAAGCA
	GIG	F	GGGCTCTCATGGGTGTAAGCTC
		R	TCCCTCTTTTGGTTCCGGTTTG
	APL3	F	CCAAACCGAATCTGAGATCGC
		R	TCCTTCCTCTGGCCTATCAGC
	CHS	F	TGATGGTGCCATAGACGGAC
		R	AGGATCGCTGGACCTCCAGG
HXK1	F	AGTGAGTGTATGGAGAGCTCAC	
	R	AGAGTGAGAAGCAGCAAGAAGCG	
ABI4	F	TCGTTTCATCATGAGGTGGCG	
	R	CGGATCCAGACCCATAGAAC	
ABI3	F	CAATGGGCTCCAAGAAGGTG	
	R	TCTCTTCGTAGCTGCTGACG	
ABI5	F	TGATCAAGAACCGCGAGTCTGC	
	R	TGTGCCCTTGACTTCAAACCTCTC	

Supplemental Figure S1.



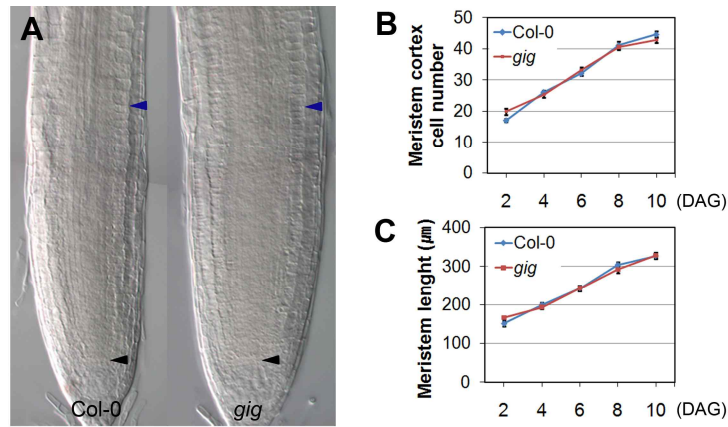
Supplemental Figure S1. Analysis of the stem cell niche in Col-0 and *gig* roots. Cell type-specific markers were used to monitor defects in and around the stem cell niche. A, *pSHR::SHR-GFP*. B, *pSCR::GFP-SCR*. C, *QC25::GUS*. D, *pWOX5::GFP*.

Supplemental Figure S2.



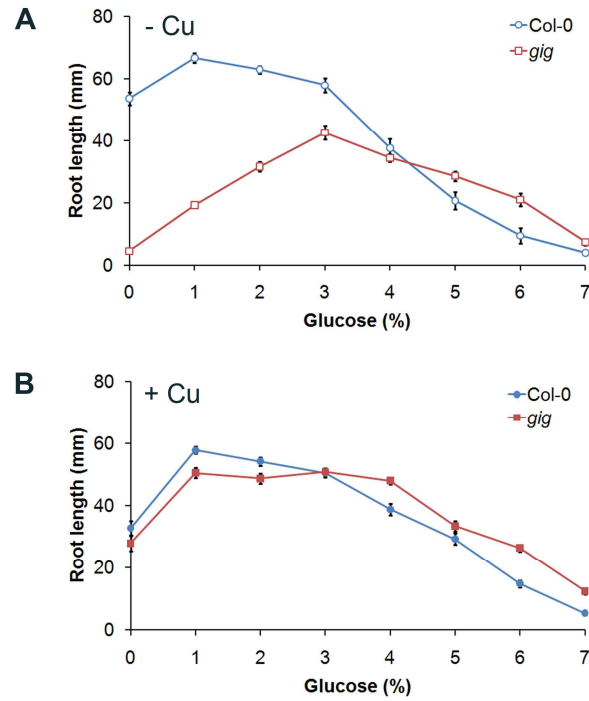
Supplemental Figure S2. Comparative analysis of Col-0 and *gig* adult plants. A, Top view. B, Side view. Rosette leaves of *gig* are smaller and inflorescence stems of *gig* are shorter than those of Col-0. F₁ progeny of crosses between Col-0 and *gig* (Col-0 x *gig*) are indistinguishably similar to Col-0.

Supplemental Figure S3.



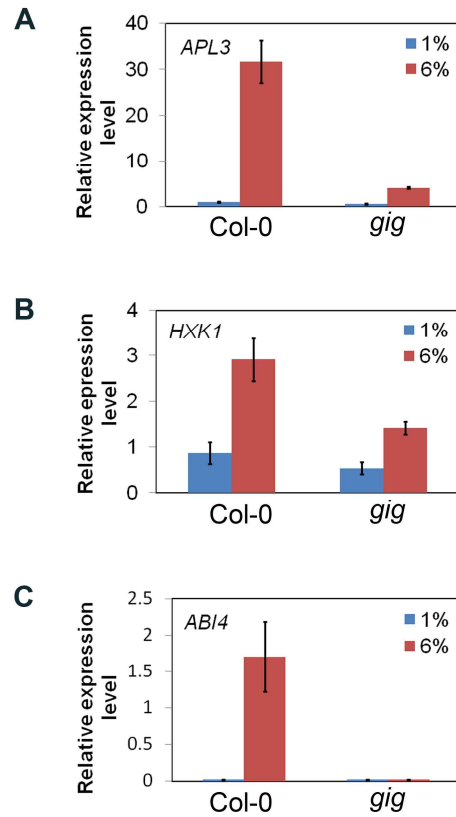
Supplemental Figure S3. Complementation of *gig* with Cu supplementation. A, DIC image of root meristem size of Col-0 and *gig* at 8 DAG in the presence of 10 µM CuSO₄. Black arrowheads indicate the QC, whereas blue arrowheads demarcate the upper border of the MZ. Measurement of meristem cortex cell number (B) and meristem length (C) from 2 to 10 DAG in the presence of 10 µM CuSO₄.

Supplemental Figure S4.



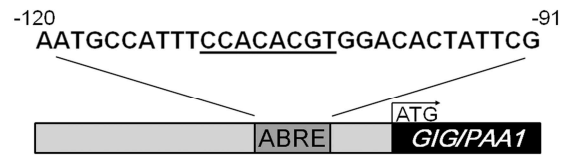
Supplemental Figure S4. Root growth assay in the absence or presence of Cu with increasing Glc concentrations. Twelve-day-old seedlings of Col-0 (blue) and *gig* (red) were grown on MS agar plates without (A) and with 10 μ M CuSO₄ (B). Error bars indicate the SE of mean from biological triplicates.

Supplemental Figure S5.



Supplemental Figure S5. Expression analysis of Glc-responsive genes with the addition of Cu. qRT-PCR in Col-0 and *gig* in the presence of 1% (blue) and 6% (red) Glc with 10 μ M CuSO₄. A, *APL3*. B, *HXK1*. C, *ABI4*. Statistical significance of differences was determined by Student's *t*-test (asterisk for $P < 0.05$). Error bars indicate the SE of mean from biological triplicates.

Supplemental Figure S6.



Supplemental Figure S6. Prediction of an ABRE sequence in the *GIG* promoter. The combination of a CCAC/ACGT core element that is known for both ABI4 binding and retrograde signaling is underlined.