

Figure S1

Supplementary Fig. S1 Phylogenetic relationship of the SAC51 family and the most related bHLH proteins in Arabidopsis. Information on bHLH proteins is based on Heim et al. (2003) and Toledo-Ortiz et al. (2003). The phylogenetic tree was constructed using the ClustalW software (<http://clustalw.ddbj.nig.ac.jp/top-j.html>). Scale bar indicates the number of nucleotide substitutions per site per year.

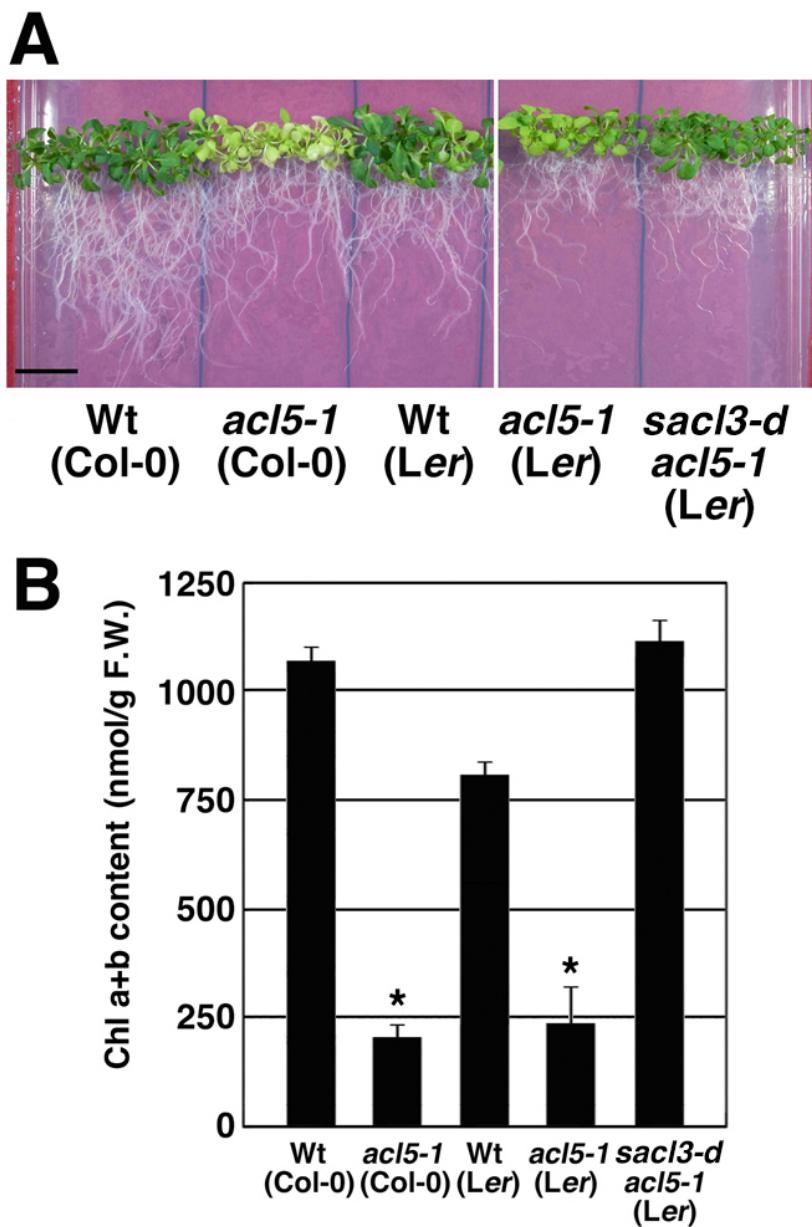


Figure S2

Supplementary Fig. S2 *sacl3-d* suppresses chlorosis in *acl5-1*. (A) Appearance of the seedlings of the wild type, *acl5-1*, and *acl5-1 sacl3-d*, grown vertically for 20 days on the MS agar plate. Scale bar: 1 cm. (B) Chlorophyll content in wild-type, *acl5-1*, and *acl5-1 sacl3-d* seedlings grown for 20 days on the MS agar plate. Values are mean \pm SD. ($n = 3$). Asterisks indicate significant differences from the wild type (* $P < 0.05$ by Student's *t*-test).

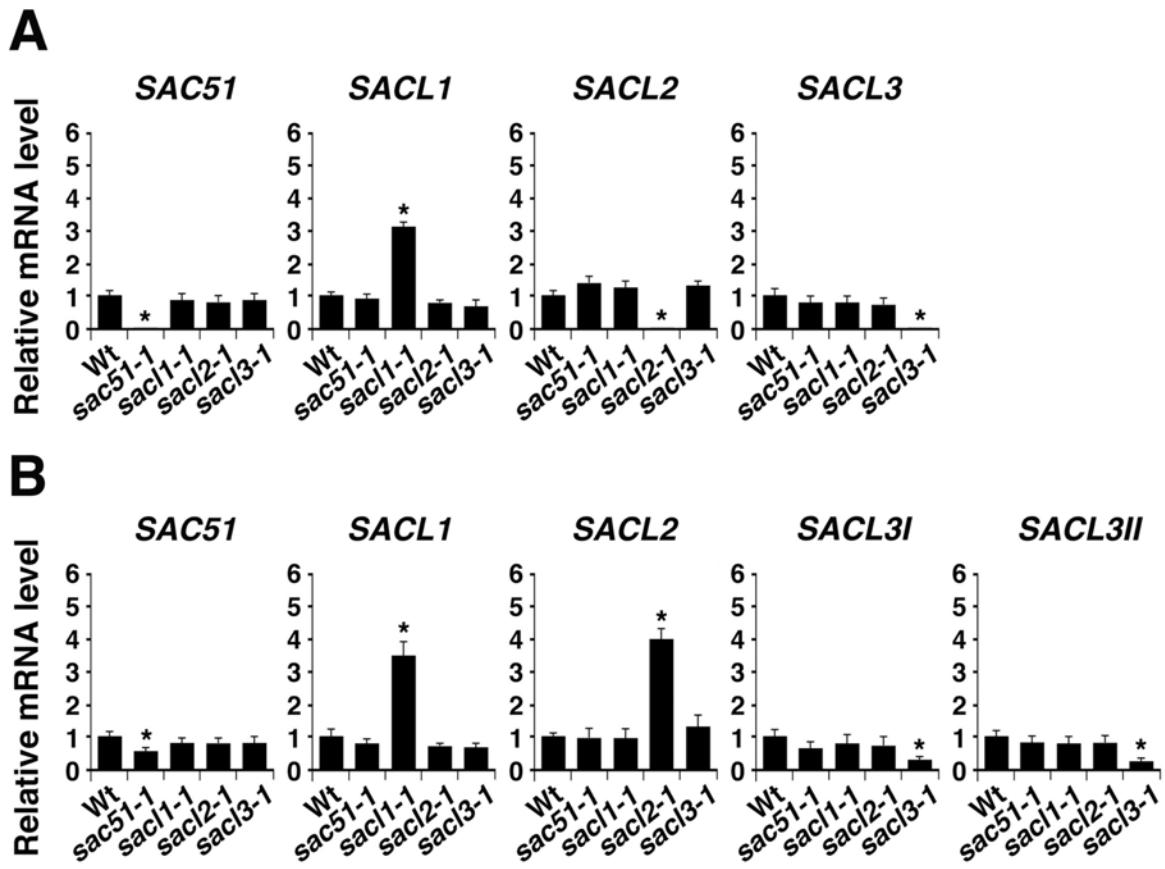


Figure S3

Supplementary Fig. S3 Expression of *SAC51* family genes in each single mutant of the family.
 (A) Relative mRNA levels of each mORF-coding region in 7-day-old seedlings of the wild type, *sac51-1*, *sac11-1*, *sacl2-1*, and *sacl3-1*. (B) Relative mRNA levels of each 5' leader region in 7-day-old seedlings of the wild type, *sac51-1*, *sac11-1*, *sacl2-1*, and *sacl3-1*. Locations of primer pairs used for RT-PCR are shown in Figure 2A. *SACL3I* and *SACL3II* represent two alternatively spliced isoforms of *SACL3*. All mRNA levels are relative to those in wild-type seedlings. Values are mean \pm SD. (n = 3). Asterisks indicate significant differences from the wild type (*P < 0.05 by Student's t-test).

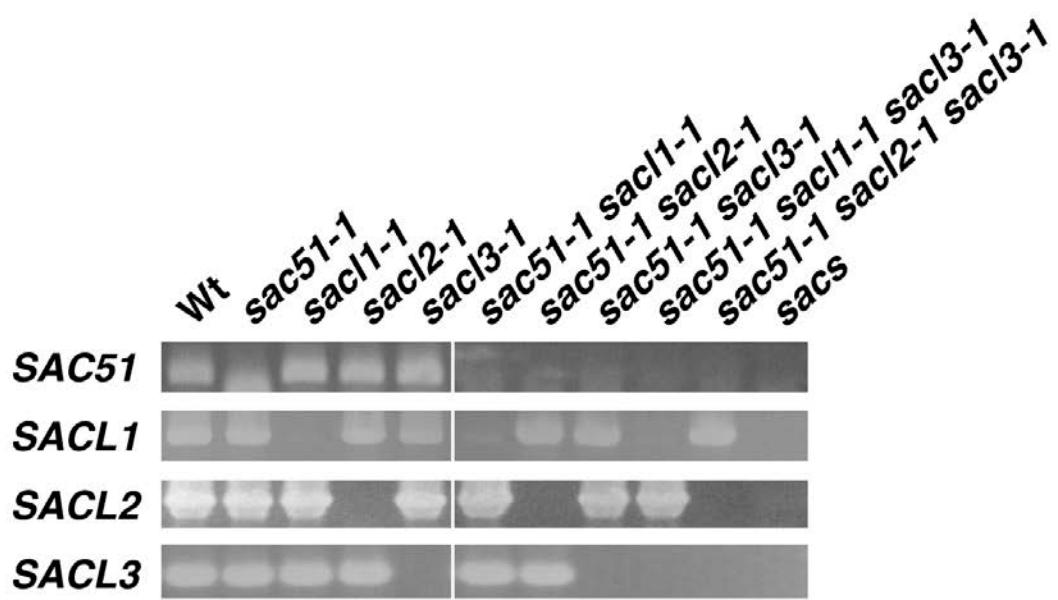


Figure S4

Supplementary Fig. S4 Confirmation by PCR of genotypes of single and multiple T-DNA insertion mutants of the *SAC51* family. Primers used for PCR are shown in Supplementary Table S1.

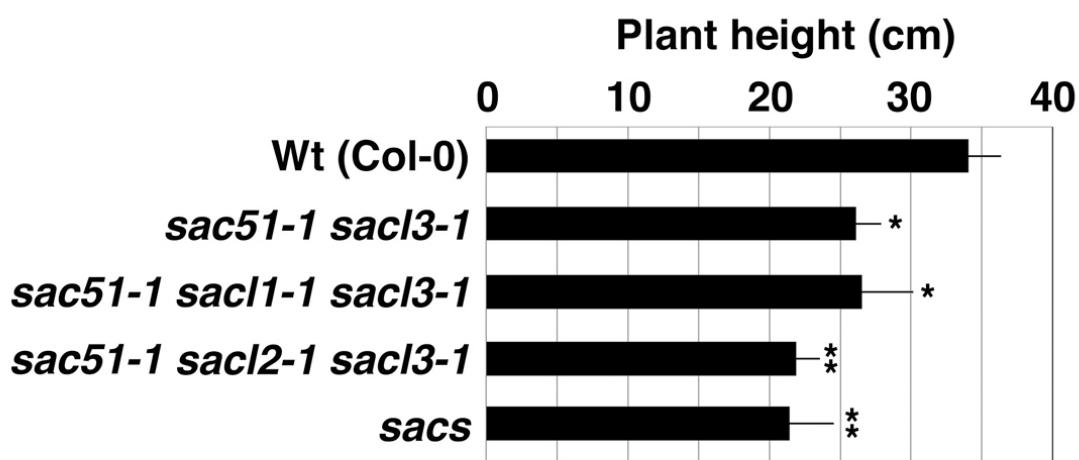


Figure S5

Supplementary Fig. S5 Comparison of plant height of the wild type (Wt (Col-0)) and each mutant grown for 40 days. Values are mean \pm SD. ($n = 8$). Asterisks indicate significant differences from the wild type (* $P < 0.05$; ** $P < 0.01$ by Student's *t*-test).

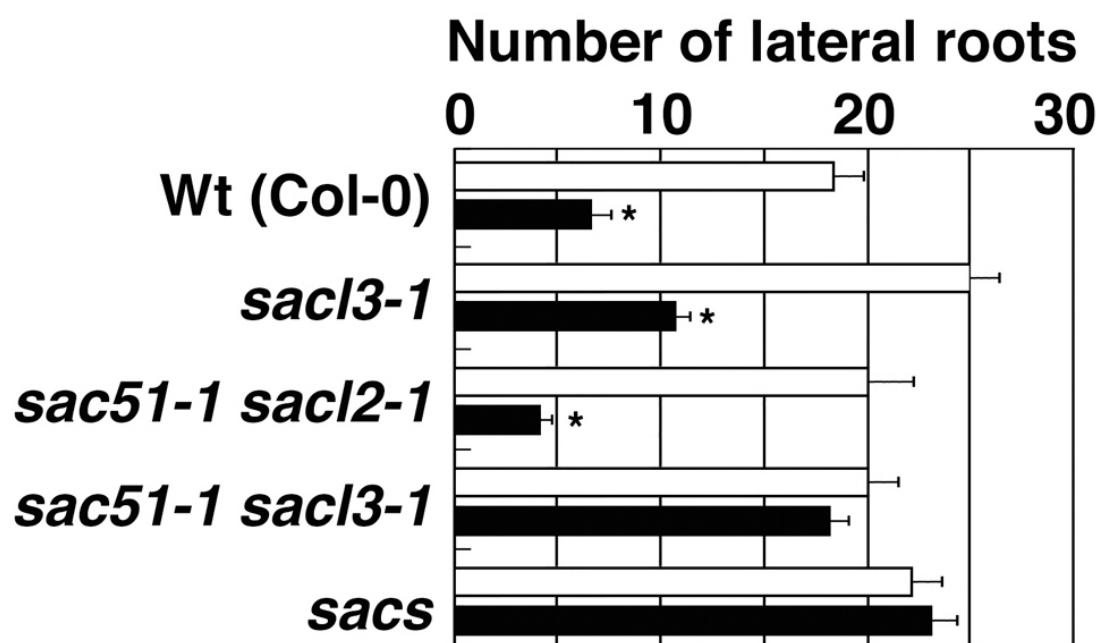


Figure S6

Supplementary Fig. S6 Effect of thermospermine on the number of lateral roots. All seedlings were grown for 10 days on the MS agar plate with no or 100 μM thermospermine. Values are mean \pm SD. ($n = 5$, $*P < 0.05$). Asterisks indicate Student's t -test significant differences.

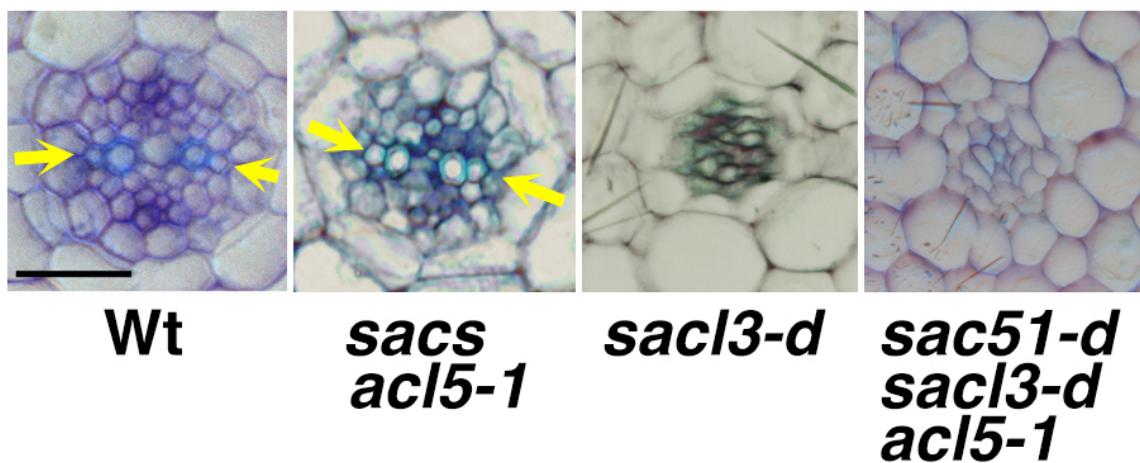


Figure S7

Supplementary Fig. S7 Vascular phenotype in the root of the wild type, *sacs acl5-1*, *sacl3-1*, and *sac51-d sacl3-d acl5-1*. Plants were grown for 10 days on the MS agar plate. Arrows indicate xylem vessel elements. Scale bar: 20 μm .

Supplementary Table S1 List of primers used for genotyping.

Primer name	Primer sequence	Enzyme for CAPS	Fragment length (bp)
ACL5dCAPS-F	5'-GGAGGTGAAGGCTCTGCTGCTCGA-3'	Xhol	WT: 215, 25
ACL5dCAPS-R	5'-TTTGTACAGAAAGCATCGCTGTTAAC-3'		<i>acl5-1</i> : 240
SAC51dCAPS-F	5'-GATTCCTCAAAGTTATGGGTAC-3'	Kpn I	WT: 105, 24
SAC51dCAPS-R	5'-TATCCATCGGTTGATAGCATTTC-3'		<i>sac51-d</i> : 129
SAC57CAPS-F	5'-TTGGTTGTGATCATGAAATCG-3'	Afl II	WT: 1400
SAC57CAPS-R	5'-ATCAGAAAACCCGTGCAAACGTA-3'		<i>sac57-d</i> : 990, 410
sac51-1F	5'-TGTCGTTGGTCACTTCATCT-3'		WT: 1300
sac51-1R	5'-TCTTAGCAGTTAGCTCAAGG-3'		<i>sac51-1</i> : 500 (R+pBI-LB)
sac1-1F	5'-GCCCACTTTAGTGAATTGC-3'		WT: 1400
sac1-1R	5'-CATGCTCTGCAGTTAACG -3'		<i>sac1-1</i> : 500 (R+Syn-LB)
sac1-2F	5'-CTCCTTAGAACTTCTATTGC-3'		WT: 1500
sac1-2R	5'-AACCTCATCTTCTTCAGAGC-3'		<i>sac1-2</i> : 400 (R+EN4-LB)
sac1-3F	5'-TCTATTGAATGTTCTGTCTGTT-3'		WT: 1800
sac1-3R	5'-GAAGAGGATACTTCTTGCTC-3'		<i>sac1-3</i> : 500 (R+pBI-LB)
pBI-LB	5'-AACCAGCGTGGACCGCTTGCTG-3'		
Syn-LB	5'-AATGGATAAAATAGCCTTGCTTCC-3'		
EN4-LB	5'-ATAACGCTGCGGACATCTAC-3'		

Supplementary Table S2 List of primers used for Ti-plasmid construction.

Primer name	Primer sequence	Purpose
SACL1-proFCI	5'-ATCGATTATAGAAACCACCAAAATGG-3'	<i>SACL1</i> promoter-5'-GUS
SACL1-proRXb	5'-TCTAGAGCAGTTAAGCCAAGGG-3'	
SACL2-proFHin	5'-GTACCGTTCCCTACTTCTATAG-3'	<i>SACL2</i> promoter-5'-GUS
SACL2-proRBm	5'-GGATCCAACACTAAACAAATCAACTA-3'	
SACL3-proFCI	5'-ATCGATTGGCACAACTTGGAAC-3'	<i>SACL3</i> promoter-5'-GUS
SACL3-proRBg	5'-AGATCTAAAAAAGGAAACGCAAAGAG-3'	

Supplementary Table S3 List of primers used for qRT-PCR.

Gene name	AGI Code	Primer sequence
<i>ACL5</i>	At5g19530	F: 5'-ACCGTTAACCAAGCGATGCTT-3' R: 5'-CCGTTAACTCTCTCTTGTTC-3'
<i>BUD2/SAMDC4</i>	At5g18930	F: 5'-ATGGCAGTGTCTGGGTTCGA-3' R: 5'-CTATTCCGACGAGGCGTGA-3'
<i>MP/ARF5</i>	At1g19850	F: 5'-GATGATCCATGGGAAGAGTT-3' R: 5'-TAAGATCGTTAATGCCTGCG-3'
<i>TMO5</i>	At3g25710	F: 5'-GAGTCATAGTGAAGCTGAGA-3' R: 5'-TCAGCTTGAGAGTTCGAAG-3'
<i>ATHB8</i>	At4g32880	F: 5'-AGCGTTTCAGCTAGCTTTGAG-3' R: 5'-CAGTTGAGGAACATGAAGCAGA-3'
<i>LOG4</i>	At3g53450	F: 5'-GCAGTTGCAGATATGCATCA-3' R: 5'-AGGCGAATATTCTCCAGCT-3'
<i>LAC6</i>	At2g46570	F: 5'-TCGAGTATGGGAGCAGGATA-3' R: 5'-ATAACCAGAGCCCTGGATTG-3'
<i>SAC51</i>	At5g64340	F: 5'-TTCTACTCAAACCAGTCACG-3' R: 5'-GTAGTAGTAGTAGTCAGC-3' (F: 5'-CATTCTTCTAAGATACTAAAG-3') (R: 5'-TTTGTGAAAGGAGAGTGAGC-3')
<i>SACL1</i>	At5g09460	F: 5'-CAGCTCCACAATCGTATTG-3' R: 5'-TGTAACAAACGAGTCTGCTC-3' (F: 5'-GAAATAATATTCCGCTTCTCG -3') (R: 5'-GATGGGTTTTCAAGGCAG-3')
<i>SACL2</i>	At5g50010	F: 5'-GGTAGTTGTGACAACACAAG-3' R: 5'-ACAACCTGATCCCTTTGTAG-3' (F: 5'-TTGCCAGGCTGAATATTCC-3') (R: 5'-TAGAAAACATGTCGGTGGTG-3')
<i>SACL3</i>	At1g29950	F: 5'-GTGGTGATTCAAGAAGAAGTC-3' R: 5'-AATAGATACCATCCTCCGA-3' (IF: 5'-CCTTCAGGTGTTGTTCCG-3') (IR: 5'-CCACAGACAGCTTCAAGAA-3') (IIF: 5'-GATTCTTCTACGAGTTGTGAG-3') (IIR: 5'-CCTGCAAAACAAGGAAGAGAT-3')
<i>ACT8</i>	At1g49240	F: 5'-GTGAGCCAGATCTCATTGTC-3' R: 5'-TCTCTTGCTCGTAGTCGACAG-3'

Primer sequences in parentheses indicate those used for amplifying 5' leader regions of each transcript (see Figure 2A).