SUPPLEMENTAL MATERIAL

Control of inflorescence architecture in tomato by BTB/POZ transcriptional regulators

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Supplemental Figure S1. *E.coli*. recombinantly expressed proteins and Western blots for GST-pull down assay. (*A*) Coomassie gel showing purified recombinant expressed His-TMF and GST-SIBOP1/2/3 proteins. (*B*, *C*) Un-cropped Western blotting images for pull down assay. Red stars note recombinant expressed proteins, Red rectangles indicate the cropped portions for preparing Fig. 1D.

NPR1	MDTTIDGFADSYEISSTSFVATDNTDSSIVYLAAEQVLTGPDVSALQLLSNSFESVFDSP
AtBOP1	MSNTFEESLKSMSLDYLNLLING
AtBOP2	-MSNLEESLRSLSLDFLNLLING
S1BOP1	MSNNPEDPLRSLSLDYLNLLING
Sibori Gipopo	
SIBOPZ	-MNTLEDSLKTLSLDYLNLLING
S1BOP3	-MNTLEDSLKTLSLDYLNLLING
	•• *: * •• •• *• *• •• BTB/POZ
NDD 1	
NPRI	DDF I SDALLVLSDGREVSFIREVLSARSSFF ASALAAARKERDSINI IA
AtBOP1	-QAFSDVTFSV-EGRLVHAHRCILAARSLFFRKFFCESDPSQPGAEPANQT-GSG
AtBOP2	-QAFSDVTFSV-EGRLVHAHRCILAARSLFFRKFFCGTDSPQPVTGIDPTQHG-SVP
S1BOP1	-OAFSDVTFHV-EGHLVHAHRCVLAARSOFFRKFFCGPSSPOSGP-OLG-SVN
S1BOP2	-OAFSDVTFSV-EGRUVHAHRCILAARSLVFRKFFCGPESPGGGP-DPSVGFG
clpop2	ARECOMMENT FOR THAT AND THE PROPERTY OF A CARD AND AND A CARD AND A CARD AND AND A CARD AND AND AND AND AND AND AND AND AND AN
51b0r5	::**:: :*.: ***:**** .*:
NDD1	
NPRI	AVLELREIARDIEVGEDSVVIVLAIVISSRVRPPRGVSECADENCENVAR
AtBOPI	ARAAAVGGVIPVNSVGIEVFLLLLQFLISGQVSIVPHKHEPRSNCGDRGCWHTHCT
AtBOP2	ASPTRGSTAPAGIIPVNSVGYEVFLLLLQFLYSGQVSIVPQKHEPRPNCGERGCWHTHCS
S1BOP1	GPRDTGSPASSVVIPVNSVGYEVFLLMMQFLYSGQVSIVPQKHEPRPNCGERGCWHTHCT
S1BOP2	SPRTSTTSSSOVVIPVNSVGYEVFLLMLOFLYSGOVSIVPOKHEPRPNCGERNCWHTHCT
SIBORS	SSDBCTTSCSOUVIDVNTVCVFVFLIMLOFLYSCOVSTVDOKHEDBDNCCFDNCWHTHCT
SIBOFS	
NPR1	PAVDFMLEVLYLAFIFKIPELITLYQRHLLDVVDKVVIEDTLVILKLANICGKACMKLLD
A+BOP1	
ACDOT 1	
AtBOPZ	AAVDLALDTLAASRIFGVEQLALLTQKQLASMVEKASIEDVMKVLIASRKQDMHQLWT
S1BOP1	SAVDLALDTLSAARSFGVEQLALLTQKQLAIMVEKASIEDVMRVLIASRKQDMNQLWT
S1BOP2	SAVDLALDTLSAARSFGVEQLALLTQKQLTSMVEKTSIEDVMKVLVASRKQDMPQLWT
S1BOP3	SAVDIAL DTL SAARSEGVEOLALL TOKHLISMVEKASIEDVMKVLIASRKODMHOLWT
515010	
NPR1	RCKEIIVKSNVDMVSLEKSLPEELVKEIIDRRKELGLEV
AtBOP1	TCSYLIAKSGLPQEILAKHLPIELVAKIEELRLKSSMPLRSLMPHHHDLTSTL
A+BOD2	
ACBOFZ	
SIBOP1	TCSHLVAKSGLPPEMLAKHLPIDVVAKIEELRLKSNLARRSLMPHHHHHLDLSSSA
S1BOP2	TCSHLVAKSGLPPEILAKHLPIDVVAKIEEIRLKTSLARRSLISHHHQHDLSSTS
S1BOP3	TCSHLVAKSGLPPEILAKHLPIDVVAKIEDLRLKSSISRRSLIPHHHHNHOHOHOMSSNI
	*. ::.**.: * * ** ::* :* : Ankvrin repeat
NPRI	PRVKRHVSNVHKALDSDDIELVKLLLKEDHTNLDDACALHFAVAYCNVKTATDLLKLDLA
AtBOP1	DLEDQKIRRMRRALDSSDVELVKLMVMGEGLNLDESLALIYAVENCSREVVKALLELGAA
AtBOP2	DLEDQKIRRMRRALDSSDVELVKLMVMGEGLNLDESLALHYAVESCSREVVKALLELGAA
S1BOP1	ELEDOKTRRMRRALDSSDVELVKLMVMGEGLNLDESTALHYAVENCSREVVKALLELGAA
SIROD2	FI FOOT DEMODAL DESDUTE VEL MUMORAL NI DESTAL UVAVENCEDEVIVENT FE CAA
516072	
SIBOP3	ELEDQKIRRMRRALDSSDVELVKLMVMGEGLNLDESLALHYAVENCSREVVKALLELGAA
	.::: .:****.*:*****:: : ***:: ** :** *. : **:*. *
NDD 1	
NPRI	DVNnRN-PRGIIVENVAAMRREPGLIESLEENGASASEATLEGRIALMIANGATMAVECN
AtBOP1	DVNYPAGPTGKTALHIAAEMVSPDMVAVLLDHHADPNVQTVDGITPLDILRTLT
AtBOP2	DVNYPAGPAGKTPLHIAAEMVSPDMVAVLLDHHADPNVRTVGGITPLDILRTLT
S1BOP1	DVNFPAGPAGKTPLHIAAEMVSPDMVAVLLDHHADPNVRMLDGITPLDILRTLT
S1BOP2	NVNHPAGPAGKTPLHTASEMVSPDMVAVLLDHHADPNVRTMDGTTPLDTLOTLT
SIROD3	
515015	•** * • **•*• *••• **•• * • * * * *
NPR1	
	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAORL
A+BOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL
AtBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLRLCL
AtBOP1 AtBOP2	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLRLCL
AtBOP1 AtBOP2 SlBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAPGLTHIEPNKLRLCL
AtBOP1 AtBOP2 SlBOP1 SlBOP2	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLRLCL
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAVPGLTHIEPNKLRLCL
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3 NPR1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAPFGLTHIEPNKLKLCL
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3 NPR1 AtBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 S1BOP1 S1BOP2 S1BOP3 NPR1 AtBOP1 btBOP2	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SlBOP1 SlBOP2 SlBOP3 NPR1 AtBOP1 AtBOP2	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLRLCL
AtBOP1 AtBOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP2 SIBOP2	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtbOP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 SIBOP2 SIBOP1 SIBOP2 SIBOP3	NIPEQCKHSIKGRLCVE I LEQEDKREQI PRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtBOP1 AtBOP1 AtBOP2 SIBOP1 SIBOP3 NPR1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP2 SIBOP2 SIBOP3 NPR1 NTR1	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtBOP1 AtBOP1 SIBOP2 SIBOP3 NPR1 AtBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtBOP1 AtBOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP1 AtBOP1 AtBOP2	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
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AtBOP1 AtBOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP3 SIBOP3 NPR1 AtBOP1 AtBOP1 AtBOP2 SIBOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGSPGLTHIEPNKLRLCL SDFLFKGSDFLFKG SDFLFKGSDFLFKG SDFLFKGSDFLFKG SDFLFKGSDFLFKG SDFLFKG SDFLFKG SDFLFKG
AtbOP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 AtbOP1 AtbOP1 SIBOP3 SIBOP1 SIBOP2 SIBOP1	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtboP1 AtboP2 SIBoP1 SIBoP2 SIBoP3 NPR1 AtbOP1 AtbOP2 SIBOP2 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 AtbOP2 SIBOP3 SIBOP2 SIBOP2 SIBOP2 SIBOP2	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtboP1 AtbOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP2 SIBOP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGSPIGLTHIEPNKLRLCL SDFLFKGSDFLFKG SDFLFKGSDFLFKG SDFLFKG
AtboP1 AtboP2 SIBoP1 SIBoP2 SIBoP3 NPR1 AtbOP2 SIBoP1 SIBoP2 SIBOP3 NPR1 AtbOP2 SIBOP3 SIBOP1 SIBOP1 SIBOP2 SIBOP1 SIBOP3	NIPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKGAPFGLTHIEPNKLRLCL
AtboP1 AtBoP2 SIBoP1 SIBoP3 NPR1 AtBOP1 AtBOP1 SIBOP2 SIBOP2 SIBOP2 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP3 NPR1 AtBOP2 SIBOP3 NPR1	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtbOP1 AtBOP2 SIBOP1 SIBOP3 NPR1 AtBOP1 AtBOP2 SIBOP1 SIBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtBOP2 SIBOP1 SIBOP3 NPR1 AtBOP3 NPR1 AtBOP1	NTPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtboP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 SIBOP2 SIBOP2 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 AtbOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 AtbOP1 AtbOP1 AtbOP1 AtbOP2	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLKLCL
AtboP1 AtboP2 SIBoP1 SIBoP2 SIBoP3 NPR1 AtbOP2 SIBoP1 SIBoP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP2 SIBOP1 SIBOP3 NPR1 AtbOP2 SIBOP3 NPR1 AtbOP2 SIBOP1 SIBOP1	NTPEQCKHSLKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL -SDFLFKG
AtboP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP2 SIBOP3 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP2 SIBOP3 SIBOP2 SIBOP3 SIBOP3	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLKLCL
AtboP1 AtbOP2 SIBOP1 SIBOP3 SIBOP3 NPR1 AtbOP2 SIBOP3 SIBOP2 SIBOP1 SIBOP2 SIBOP1 AtbOP2 SIBOP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP2 SIBOP3 NPR1 AtbOP1 SIBOP3 SIBOP3	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSFAVAADELKMTLLDLENRVALAQRL SDFLFKG
AtboP1 AtbOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 SIBOP2 SIBOP3 SIBOP2 SIBOP3 SIBOP3 SIBOP2 SIBOP1 SIBOP2 SIBOP3 NPR1 AtbOP1 AtbOP1 AtbOP1 AtbOP1 SIBOP2 SIBOP3 SIBOP3	NIPEQCKHSIKGRLCVEILEQEDKREQIPRDVPPSPAVAADELKMTLLDLENRVALAQRL SDFLFKGAIPGLTHIEPNKLRLCL

Supplemental Figure S2. Alignment of SIBOP1/2/3 with NPR1 and BOP1/2 proteins from *Arabidopsis* (AtBOP1/2). Single and double lines indicate the BTB/POZ and ankyrin repeat domains, respectively. Blue color highlights the two essential domains and two oxidized cysteine residues in NPR1 that are required for its SA-induced transcriptional regulation, which are absent in SIBOP proteins. Green color and arrow noted the cysteine residue in NPR1 that is critical for its S-nitrosylation and SA-induced oligomerization. Red color highlighted the four conserved cysteine residues in the BTB/POZ domain that are shared among all six BTB/POZ proteins and are important for oligomerization of NPR1. The highly conserved domain marked by the four cysteine residues is labeled by red rectangles.



Supplemental Figure S3. Pleiotropic vegetative and reproductive phenotypes of *CR*slbop single and higher-order mutant plants. (*A-H*) Representative mature leaf from WT (*A*), single mutants of *CR*-slbop1 (*B*), *CR*-slbop2 (*C*), *CR*-slbop3 (*D*), double mutants of *CR*-slbop1/2 (*E*), *CR*-slbop2/3 (*F*), *CR*-slbop1/3 (*G*), and *CR*-slbop1/2/3 triple mutants (*H*) showing altered leaf complexity and floral organ abscission defects (insets). Inset in (*C*) shows ectopic shoot generation on the rachis of a *CR*-slbop2 leaf. Red asterisks indicate intercalary leaflets and floral abscission defects, respectively. (*I*) Quantification and statistical comparison of leaflets in WT and *CR*-slbop mutants. (*J*, *K*) Representative fruits from WT and *CR*-slbop mutants showing scars on fruits (*J*) and fruit shape phenotypes (*K*) in *CR*-slbop mutants. (*M*, *N*) Representative flowers from WT (*M*) and

CR-slbop1/2/3 triple mutants (*N*) showing ectopic twisted stigmas fused with stamens in *CR-slbop1/2/3* triple mutants. Inset shows ectopic stigma fused to the carpels of an ovary, explaining the origin of fruit scarring. (*O*) Representative leaves from single and higher order *CR-slbop* mutants grown under greenhouse conditions, where leaf phenotypes were often enhanced. For example, *CR-slbop1* leaves appeared larger than WT, primarily due to longer petiolules, and *CR-slbop2* leaves developed more intercalary leaflets that were frequently fused along the rachis. Insets show rare examples of more extreme leaves. Data are means (±s.d.); n = 5. A two-tailed, two-sample Student's *t* test was performed, and significant differences are represented by black asterisks: *P < 0.05, **P < 0.01, ***P < 0.001. Scale bars, 2 cm (*A-K*), 500 µm (*M*, *N*), 5 cm (*O*).

S1BOP1	ATGAGTAATAATCCTGAAGATCCCTTAAGAAGTCTCTCTTTAGATTATCTCAATCTCCTC
S1BOP2	ATGAATACTCTTGAAGATTCCTTAAAAACTCTCTCTTTAGATTATCTAAATCTTCTC
SIBOP3	ATGAATACTCTTGAAGATTCCTTAAAAACTCTCTCTTTTAGATTATCTAAATCTTTCT * **** ** ****** ****** ** **********
S1BOP1	ATCAATGGTCAAGCTTTCAGTGATGTTACTTTTCATGTTGAAGGTCATTTAGTCCATGCT
S1BOP2	ATCAATGGTCAAGCTTTTAGTGATGTTACTTTTAGTGTTGAAGGTCGTTTAATACATGCT
S1BOP3	ATCAACGGTCAAGCTTTTAGTGATGTTACTTTTAGTGTTGAAGGTCGTTTAGTACATGCT
	**** ********* ************************
S1BOP1	CACCGTTGCGTCCTGGCAGCAAGGAGTCAATTCTTCAGAAAATTTTTCTGCGGGCCGAGC
S1BOP2	CATAGATGCATCTTAGCTGCTAGAAGTCTCTTCTTCCGCAAATTCTTCTGCGGGCCGGAG
S1BOP3	CATAGATGCATCTTAGCTGCTAGAAGTCTTGTGTTTAGAAAATTCTTCTGCGGGCCTGAG
	** * *** ** * ** ** ** *** * ** * ******
S1BOP1	TCTCCTCAGTCCGGCCCGCAACTCGGCTCGGTTAACGGG
Slbop2	TCAGCATCTGTCTCCGGCCCGCGGTTGGGCCCGTTCGGCGTTGGCGCTGGATTAGCGTCG
Slbop3	TCGCCTGGTGGCGGTCCAGACCCTTCCGTGGGCTTTGGAAGC
	** * * *
Slbop1	CCGAGAGATACTGGTTCACCAGCATCATCAGTAGTGATACCGGTGAATTCAGTAGGATAT
S1BOP2	TCGCCGAGGGGCACAACAAGTTGTTCACAAGTAGTAATACCTGTGAACACTGTAGGGTAT
S1BOP3	CCGCGGACTAGTACTACCAGTAGTTCACAGGTAGTAATACCTGTGAACTCGGTAGGGTAT ** ** ***** ***** ***** ***** * *****
SIROP1	
SIBOP2	GAGGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTGTGGGCCACAAGTTTCTAATTGTGCCCCCACA
SIBOP3	GAGGTTTTTTTGTTGATGTTACAATTTTTGTATAGTGGACAAGTTTCAATTGTGCCACAA
	**** **** ***** * ** ***** ************
S1BOP1	AAACATGAGCCAAGGCCAAATTGTGGAGAGAGAGGGTTGTTGGCATACACATTGCACCTCA
S1BOP2	AAACATGAACCAAGGCCTAATTGTGGTGAAAGAAACTGTTGGCATACACATTGCACATCA
S1BOP3	AAACATGAACCAAGGCCTAATTGTGGTGAAAGAAACTGTTGGCATACACATTGCACATCA
Slbop1	GCCGTTGATCTTGCACTTGATACACTCTCAGCCGCTAGATCTTTTGGTGTTGAACAACTT
Slbop2	GCCGTTGATCTTGCACTGGATACACTCTCAGCCGCTAGATCTTTTGGAGTTGAACAACTT
S1BOP3	GCCGTTGATCTTGCACTTGATACACTCTCAGCCGCTAGATCTTTTGGTGTTGAACAACTT *******************************
S1BOP1	GCTTTGCTCACTCAGAAGCAATTGGCAATCATGGTAGAAAAAGCTTCAATTGAGGATGTG
S1BOP2	GCTTTGCTTACTCAGAAGCATTTGATAAGCATGGTGGAAAAAGCTTCAATTGAGGATGTG
S1BOP3	GCTTTGCTTACTCAGAAGCAATTGACAAGCATGGTAGAGAAAACTTCAATTGAGGATGTG ******* *********** *** ** ****** ** **
Slbop1	ATGAGAGTTCTAATAGCATCAAGGAAGCAAGACATGAATCAACTATGGACTACGTGTTCA
Slbop2	ATGAAAGTTTTAATAGCTTCAAGAAAACAAGACATGCATCAACTTTGGACTACTTGTTCT
Slbop3	ATGAAAGTTTTAGTTGCTTCAAGAAAACAAGACATGCCTCAACTTTGGACTACTTGTTCT **** **** ** * * * ***** ** ******** ****
Slbop1	CATTTGGTTGCAAAATCAGGTCTTCCACCCGAAATGTTGGCCAAACACCTCCCCATTGAT
S1BOP2	CATTTGGTTGCTAAATCAGGTCTCCCACCTGAAATCCTAGCCAAACACCTCCCTATTGAT
S1BOP3	CATTTGGTTGCAAAATCAGGCCTTCCACCTGAAATCCTAGCAAAACACCTTCCCATCGAC
S1BOP1	GTTGTAGCCAAAATTGAAGAACTACGCCTCAAATCCAACCTAGCACGTCGATCCTTAATG
S1BOP2	GTTGTTGCCAAAATCGAAGACCTTCGTCTCAAATCCTCTATATCGAGAAGATCTTTAATC
S1BOP3	GTTGTCGCGAAAATCGAGGAAATTCGCCTAAAAACTTCATTAGCGCGAAGATCCTTGATC ***** ** ***** ** ** * ** ** ** ** ** *
SlBOP1	CCACATCATCATCACCACCTCGACCTCAGCTCTCAGCTGAGCTCGAG
S1BOP2	CCTCATCATCATCACAACCATCAACATCAACATCAAATGTCCTCGAACATTGAGCTCGAG
S1BOP3	TCTCACCATCACCAGCACGACCTAAGTTCGACCTCGGAGCTCGAG * ** ***** ** ** ** ** ** ** ** ** ** *
S1BOP1	GACCAAAAAATCCGTAGGATGAGACGAGCCCTTGACTCATCGGACGTTGAACTTGT
S1BOP2	GACCAGAAGATCCGACGAATGAGACGAGCGCTCGATTCGTCGGACGTGGAATTAGT
S1BOP3	GACCAAAAGATCCGACGAATGAGACGAGCCCTAGACTCATCCGACGTGGAATTGGT
	***** ** ***** * ********* ** ** ** **

Supplemental Figure S4. Alignment of *SlBOP1, 2, 3* coding sequences used for generating the hairpin RNAi construct to knockdown *SlBOP1, 2* and *3* transcript levels in *RNAi-slbop* plants. Note the stretches of nucleotide consensus among all three genes, indicated by red stars.



Supplemental Figure S5. Molecular and phenotypic analysis of *RNAi-slbop* plants. (*A*) qRT-PCR showing transcriptional knock-down of *SlBOP1*, 2 and 3 at the TM stage of *RNAi-slbop #1* used for genetic analyses shown in Figure 6. (*B*) Quantification and comparison of flowers per inflorescence in WT and *RNAi-slbop #1* plants. (*C*) Representative mature leaf from *RNAi-slbop #2* plants. (*D*) Quantification of flower number per inflorescence from an additional *RNAi-slbop* transgenic line. Data are means $(\pm s.d.)$; n = 3 (*A*), 12 (*B*, *D*). A two-tailed, two-sample Student's *t* test was performed, and significant differences are represented by black asterisks: ****P* < 0.001. Scale bar, 5 cm.



Supplemental Figure S6. Inflorescence phenotypes of *RNAi-slbop an* and *RNAi-slbop fa* mutant plants. (*A*, *B*) A representative inflorescence for *an* mutant (*A*) and *RNAi-slbop an* mutant plants (*B*). *RNAi-slbop an* plants branch less frequently than *an* mutants, as measured by the first branches, and thus SIMs, produced during early inflorescence development (insets and bar graph). (*C*, *D*) A highly branched leafy inflorescence from *fa* mutant (*C*) and *RNAi-slbop fa* mutant plants (*D*). Data are means (±s.d.); n = 21. A two-tailed, two-sample Student's *t* test was performed, and significant differences are represented by black asterisks: **P < 0.01. Scale bars, 2 cm.



Supplemental Figure S7. *RNAi-slbop* enhances a weak allele of *tmf*. (*A*) Representative primary shoot of the weak *tmf-2* mutation that flowers early, but shows normal multiflowered primary inflorescences. (*B*) Introducing *RNAi-slbop* into the *tmf-2* background by crossing results in earlier flowering compared to *tmf-2* alone, and nearly full penetrance for single-flower primary inflorescences that also exhibit enhanced leaf-like sepals. (*C*) A whole plant view of *RNAi-slbop tmf-2* plants showing enhanced early flowering and single-flower inflorescences in both primary and side shoots showing leaf-like sepals. Red arrows indicate inflorescences. Leaves were removed to show inflorescence phenotype in *tmf-2* and *RNAi-slbop tmf-2* plants. L: leaf. Scale bars: 2.5 cm (*A*, *B*), 5 cm (*C*).



Supplemental Figure S8. CRISPR/Cas9-generated null mutations in *TFAM1* and 2 result in phenotypes that match multiple defects of *slbop* single mutants. (*A*) Normalized RNA-seq read counts for *TFAM1, 2,* and 3 in five stages of primary shoot meristem maturation showing dynamic expression of *TFAM1* and *TFAM2*, similar to *TMF* and *SlBOP1* and 2 (Fig.3). (*B*) Yeast two-hybrid assays showing SlBOP1, 2, and 3 interact

with TFAM1 and TFAM2. (C) Schematics illustrating two sgRNAs (red arrows) targeting TFAM1 and TFAM2. Black arrows represent PCR genotyping primers. (D) Sequences from homozygous T1 transgenic progeny plants lacking the Cas9 transgene showing out-of-frame deletions (blue dashed lines) for *CR-tfam1* and 2, resulting in two independent null loss-of-function alleles (a1, a2). Red font highlights sgRNA targets, and black boxes indicate PAM sequences. (E) Quantification and statistical comparison of leaflets in WT and CR-tfam1 and 2 mutants. (F, G) Representative inflorescences (red arrow) from CR-tfam1 (F) and CR-tfam2 mutants (G). Red arrowhead in (G) indicates a single branching event often observed in *CR-tfam2* mutants, similar to *CR-slbop2* mutants (Figure 4D). (H) Quantification and statistical comparison of flowers per inflorescence in WT and CR-tfam1 and 2 mutants. Note that flower number for CRtfam2 plants was quantified as the flowers on each inflorescence branch from branched inflorescences, and from unbranched inflorescences separately. (1) Representative flowers from *CR-tfam1* (left), *CR-tfam2* (right) mutants showing ectopic twisted stigmas fused with stamens (arrowheads) in CR-tfam1. (J, K) Representative fruits from CR-tfam1 and 2 showing floral organ abscission defects (J), scars on fruits, and altered fruit shape (K) in *CR-tfam1*, similar to *CR-slbop* single mutants (Supplemental Fig. S3). Scale bars, 2 cm (F, G, K), 500 μ m (I, J). Data are means (±s.d.); n = 5-11 (E); n = 11-21 (H). A twotailed, two-sample Student's t test was performed, and significant differences represented by black asterisks: *P < 0.05, ***P < 0.001.



Supplemental Figure S9. Driving expression of the flower specification gene *AN* in the earliest stage of meristem maturation using *TMF* promoter (Transactivation, LhG4, OP system) results in single-flower primary inflorescence after producing only two leaves. Scale bars, 2 cm.

	Guide RNA	A sequences	Genotyping	g primers
Gene	Target 1	Target 2	Forward	Reverse
SIBOP1	ATTGGGCCGAGCTCTCCTCAGTC	ATTGTGATACCGGTGAATTCAGT	CACAAGACAAAACACCAACCA	GCTCATGTTTTTGCGGTACA
SIBOP2	ATTGGCCCGCGGTTGGGCCCGTT	ATTGTAATACCTGTGAACACTGT	GCCACTAACCTTTCTCCTATTG	GGCTGATGTGCAATGTGTATG
SIBOP3	ATTGGGCCTGAGTCGCCTGGTGG	ATTGGTAGTAATACCTGTGAACT	CACAAAATCATCTTCCACTATGTGA	TGCAATGTGTATGCCAACAG
TFAM1	ATTGCCGCCTTCGTCACCACCGA	ATTCTGATACTTGCGCGATGCA	AAAGTAACACAGCCATGTTAGACG	GCCGAAAAACGGACAATTAC
TFAM2	ATTGGAAATAATTTCACTACATC	ATTGTGCACTAGTGCACATATCC	TTTCCTCCCTATTTCAAGAATCA I	CCTTCAAAAATTACCACTTTATCC

Supplemental Table S1. Single guide RNA (sgRNA) sequences and genotyping primers

of CR-slbops and CR-tfams.

Primer name	Primer Sequence 5' to 3'
SIBOP1-F	TGGCATCACTCCATTGGACA
SIBOP1-R	GATCAATGTTTGCGCTCCCT
SIBOP2-F	GGGTGAAGCGAATAACAATCAA
SIBOP2-R	TGCATGCCATTTGATGATGATG
SIBOP3-F	TCTTCCACCGCGATATTCCA
SIBOP3-R	TTCGTCATTCTGTTGCACCC
FA-F	CACTGCCTTGATGAGGATGC
FA-R	TCAGAATGGCACAGCTGTCG
AN-F	GAGCCTTGTTGAAGGGAATG
AN-R	CCTTGTCCATTCTCCACTTC
SISEP3-F	CGGCAAACAACTCAAACTCA
SISEP3-R	TGATAGGAAAACCATTGAGCA
FUL-F	AGAAAGCGCTCCAAGAACAA
FUL-R	CCGATCCATCTCTGTTCATTCC
S-F	TTCAACTGTTTCTCCTCTTGCT
S-R	CCTATCCCAACCTCAAAAGC

Supplemental Table S2. qRT-PCR primers used in this study.