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Supplementary information

A spatiotemporal molecular switch governs plant asymmetric cell division

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Supplementary Information for

A Spatiotemporal Molecular Switch Governs Plant Asymmetric Cell Division

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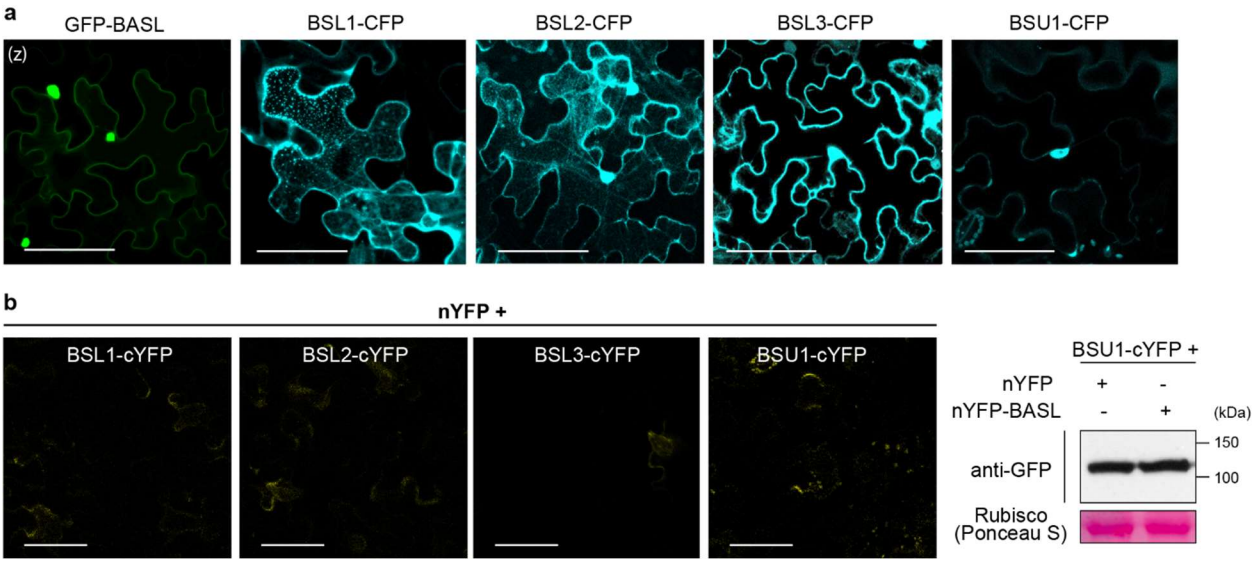
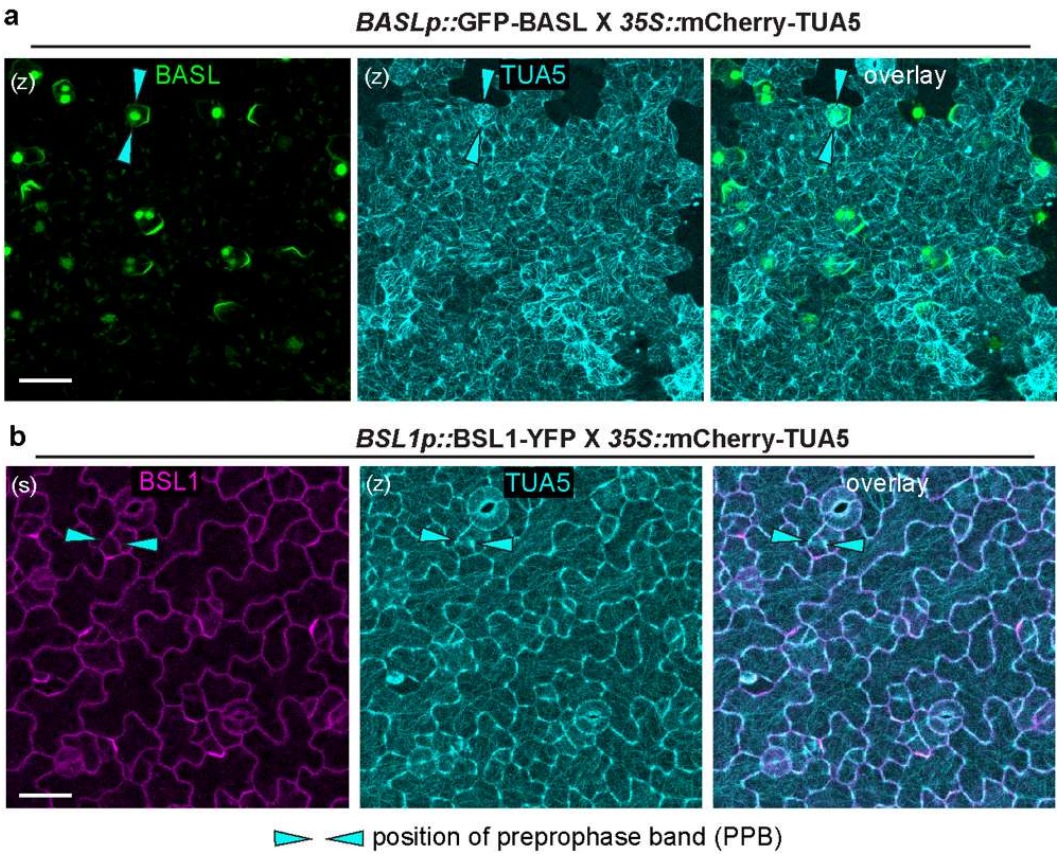


Supplementary Fig. 1 | BSL1, BSL2 and BSL3 Physically Interact with BASL *in Planta*
a, Representative confocal images show subcellular localization of indicated proteins in *N. benthamiana* leaf epidermal cells. BASL (green), nuclear and cytoplasmic; BSL1 (cyan), cell membrane and cytoplasmic puncta; BSL2 (cyan), nuclear and cytoplasmic; BSL3 (cyan), nuclear and cytoplasmic; BSU1 (cyan), predominantly nuclear and weakly cytoplasmic. (z), z-staked confocal images.
b, Negative controls for Fig. 1c BiFC assays testing BSL-BASL interaction in *N. benthamiana* leaf epidermal cells. Confocal images show no significant YFP signals were recovered when nYFP was coupled with BSL-cYFP fusions. nYFP and cYFP, N- and C-terminal domain of the split YFP, respectively. Western blots show BSU1-cYFP protein was detected with anti-GFP when BSU1-cYFP co-expressed with nYFP or nYFP-BASL in *N. benthamiana* leaves. Data represent results of experiments repeated three times. Protein loading is shown by Ponceau S staining (bottom). Scale bars in (**a-b**), 50 μm.



Supplementary Fig. 2 | Determination of Polarization Timing for BASL and BSL1
a-b, Co-expression of the native promoter driven GFP-BASL (green)(**a**) or BSL1-YFP (magenta)(**b**) with the microtubule marker mCherry-TUA5 (cyan, driven by the ubiquitous *35S* promoter) in 60-hpg adaxial cotyledon epidermis in *Arabidopsis*. The expression of the microtubule marker mCherry-TUA5 allows the visualization of the preprophase band (PPB) (cyan arrowheads) that forms at the G2 phase of the cell cycle. Scale bar, 25 μm. (z), images are z-projected. (s), single optional sections.

Supplementary Tables

Table S2. List of primers used in this study
The name and sequence of these primers was displayed in left. The purposes of these primers were listed in right. F, forward; R, reverse.

Name	Primers	Purpose
BSL1-F-NotI	GCTCCGCGGCCGCC ATGGGCTCGAAGCCTTG	for pENTR-D1/BSL1-CDS and genomic,overexpression lines, Y2H, BiFc
BSL1-R-AscI(-stop)	A GGCGCGCCC GATGTATGCAAGCGAGCTTCTG	
BSL2-F-NotI	GCTCCGCGGCCGCC ATGGATGAAGATTCGTCTATGG	for pENTR-D1/BSL2-CDS and genomic,overexpression lines, Y2H, BiFc
BSL2-R-AscI(-stop)	A GGCGCGCCC CATCCAAGCCAGAGAACC	
BSL3-F-NotI	GCTCCGCGGCCGCC ATGGATTTGGATTCTTCAATG	for pENTR-D1/BSL3-CDS and genomic,overexpression lines, Y2H, BiFc
BSL3-R-AscI(-stop)	A GGCGCGCCC TATCCAAGCAAGAGAGC	
BSU1-F-NotI	GCTCCGCGGCCGCC ATGGCTCCTGATCAATCTTATC	for pENTR-D1/BSU1-CDS and genomic,overexpression lines, Y2H, BiFc
BSU1-R-AscI(-stop)	A GGCGCGCCC TTCACTTGACTCCCCTC	
BSL1 promoter-F	GCTCCGCGGCCGCC ACTCAGTTGCATTGAATTTGAC	for pENTR-D1/BSL1promoter-genomic
BSL1 promoter-R	GCTCCGCGGCCGCC TGGAAACCACTTTACGGGTATAAATC	
BSL2 promoter-F	GCTCCGCGGCCGCC TTATCAAATTGTAGTCCATCCAAG	for pENTR-D1/BSL2promoter-genomic
BSL2 promoter-R	GCTCCGCGGCCGCC TATCAAAAAGCTTCAAAAGTGG	
BSL3 promoter-F	GCTCCGCGGCCGCC TTCGGTCTTGATGGAACG	for pENTR-D1/BSL3promoter-genomic
BSL3 promoter-R	GCTCCGCGGCCGCC ATTTTTCACAACCCTAAATTCGTC	
BSU1 promoter-F	GCTCCGCGGCCGCC AAACCACTGACATCTCTTCATC	for pENTR-D1/BSU1promoter-genomic
BSU1 promoter-R	GCTCCGCGGCCGCC AACACAATATTTTGTGGTGG	
BIN2cds-F-NotI	GCTCCGCGGCCGCC ATGGCTGATGATAAGGAGATG	for overexpression lines, BiFc
BIN2pro-F-NotI	GCTCCGCGGCCGCC CTCGGTTATACAATGAGGTTATC	for pENTR-D1/BIN2promoter-genomic
BIN2-R-AscI(-stop)	A GGCGCGCCC AGTTCCAGATTGATTCAAGAAG	
BSL1D584N-F	CCAAATTGCCCATGGAGATTGCCAAATACTTTGATAGGA	point mutation

BSL1D584N-R	TCCTATCAAAGTATTTGGCAATC TCCATGGGCAATTTGG	
BSL1cds-F-SmaI	CC CCCGGG ATGGGCTCGAAGCCTTG	Protein fusion with GST
BSL1cds-F-XbaI	GCTCTAGA ATGGGCTCGAAGCCTTG	Protein fusion with MBP
BSL1cds-R-SalI(-stop)	GC GTCGAC GATGTATGCAAGCGAGCTTCTG	Protein fusion
BSL2cds-F-EcoRI	CG GAATTC ATGGATGAAGATTCGTCTATGG	Protein fusion with MBP, GST
BSL2cds-R-SalI(-stop)	GC GTCGAC CATCCAAGCCAGAGAACC	
BSL3cds-F-XbaI	GCTCTAGA ATGGATTTGGATTCTTCAATG	Protein fusion with MBP
BSL3-R-SalI(-stop)	GC GTCGAC TATCCAAGCAAGAGAGC	
BSU1-F-EcoRI	CG GAATTC ATGGCTCCTGATCAATCTTATC	Protein fusion with MBP, GST
BSU1-R-SalI(-stop)	GC GTCGAC TTCACTTGACTCCCCTC	
MBP-YDA F	C GAGCTC ATGCCTTGGTGGAGTAAATC	Protein fusion with MBP
MBP-YDA R	CC AAGCTT GGGTCCTCTGTTTGTTGATC	
pET28a-BASL F	CG GAATTC ATGGCTTCACAGTGGACAATAC	Protein fusion
pET28a-BASL(-Stop) R	CC CTCGAG GAATCTACAACATTGGAACC	
BIN2-F EcoR1	CG GAATTC ATGGCTGATGATAAGGAGATG	Protein fusion with GST
BIN2cds-R-NotI(-stop)	GCTCCGCGGCCGCC AGTTCCAGATTGATTCAAGAAG	
SALK_051383(BSL1) LP	TGATTAAATCTTGTCCACGCC	Mutant genotyping
SALK_051383(BSL1) RP	GCTTCATCCGAGAGCTGTATG	
SALK_147279(BSL1) LP	GACCTCGAAACTGGAAACCTC	Mutant genotyping
SALK_147279(BSL1) RP	TAGGGGTGATTTACCCCAAAC	
SALK_055335(BSL2) LP	CATTAGCAAAGTTCTGCCAGC	Mutant genotyping
SALK_055335(BSL2) RP	GTTCCAGAGCAGATGGAGATG	
WiscDsLox245G08-LP	TGAAGATCGTTGTTGTTGCAG	Mutant genotyping
WiscDsLox245G08-RP	AAACTTGTGACATCAGTGGCC	
SALK_071689(BSL3) LP	CAAACATTTGAAAGGGTACGATG	Mutant genotyping

SALK_071689(BSL3) RP	AAAACATACGAATGCCAGCAC	
SALK_072437(BSL3) LP	CCTGCAAAATATCAATGCTTAG	Mutant genotyping
SALK_072437(BSL3) RP	TAATGCACTTTTTGGTTTCCG	
SALK_030721(BSU1) LP	ACGTTCCACTTCAACATGGAG	Mutant genotyping
SALK_030721(BSU1) RP	TCTTTAACCATGCTTCGAACC	
SAIL_101_H03 LP	TCAACAAAGGGTCCACAACTC	Mutant genotyping
SAIL_101_H03 LP	TGTCCACTTCCTGGTCAAAAC	
BSL1 C-LP	GACGACGCTTGGATGCAGGAGCTG	RT-PCR, C to 3'utr 279bp
BSL1 3'utr-RP	CTATACCATTCTCACTCTCTGGT	
BSL2-RT LP	GAAGACACATGGATGCAGGAGCT	RT-PCR, C to 3'utr 490bp
BSL2-RT RP	CACCTAATCAACCATTACCATTC	
BSL3-RT LP	AGAGGATACATGGATGCAGGAGTTA	RT-PCR, C to 3'utr 624bp
BSL3-RT RP	CAAACGACCAAACACACCTCTCT	
BSU1Ct-RT	CTCCCATCTCATCTTCAG	RT-PCR, C to 3'utr 225bp
BSU1 3'utr-R	CCTCTGCCAATACCAAAATAG	
BSL1-qPCR-R	CCTCCCTCAATAGCGGTGGCG	qPCR
BSL2-qPCR-R	GCTCTTGTCCAACAACCGCA	qPCR
BSL3-qPCR-R	CTGTTGCTGCTGTTGTTG	qPCR
BASL-qPCR-F	CTCTGATAAATCTAGCGGGTC	qPCR
BASL-qPCR-R	CTACAACATTGGAACCCTAAAG	qPCR
BIN2-qPCR-F	GTGACTTTGGCAGTGCGAAAC	qPCR
BIN2-qPCR-R	CAGCATTTTCTCCGGGAAATAATGG	qPCR