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Supplementary Information for

BAM1/2 receptor kinase signaling drives CLE-peptide mediated formative cell divisions in Arabidopsis roots

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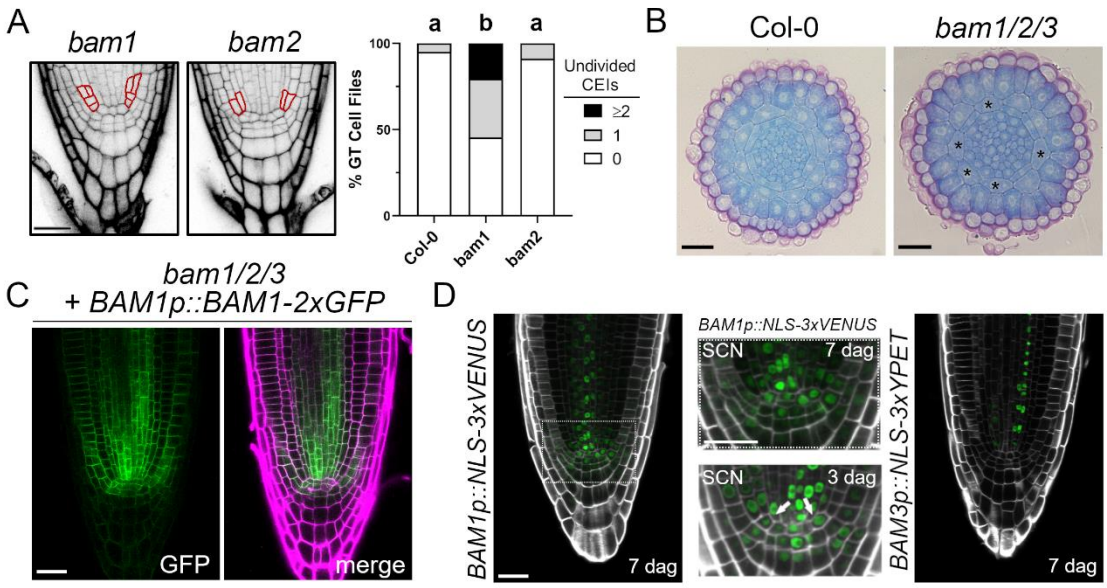

Fig. S1. BAM1/2 receptors are required for formative root cell divisions. (A) Quantification of undivided CEI cells in *bam1* and *bam2* single mutants (5 dag; n=81, Col-0; n= 56, *bam1*; n= 48, *bam2*). Undivided CEI cells and the first set of divisions are outlined in red. Distributions were compared using a Kruskal-Wallis nonparametric test. (B) Cross-sections of Col-0 and *bam1/2/3* roots at 6 dag. Asterisks represent a GT cell file with no formative division. (C) The division defects of a *bam1/2/3* mutant are rescued by the *BAM1p*::*BAM1-2xGFP* transgene. (D) The promoter of *BAM1* is active throughout the stem cell niche (SCN; boxed area), ground tissue (including CEIs at 3 dag, arrows), and stele in roots at 3 and 7 dag. (D) *BAM3* expression is restricted mainly to phloem lineage cells (D). Scale bars = 25 uM (A-D).

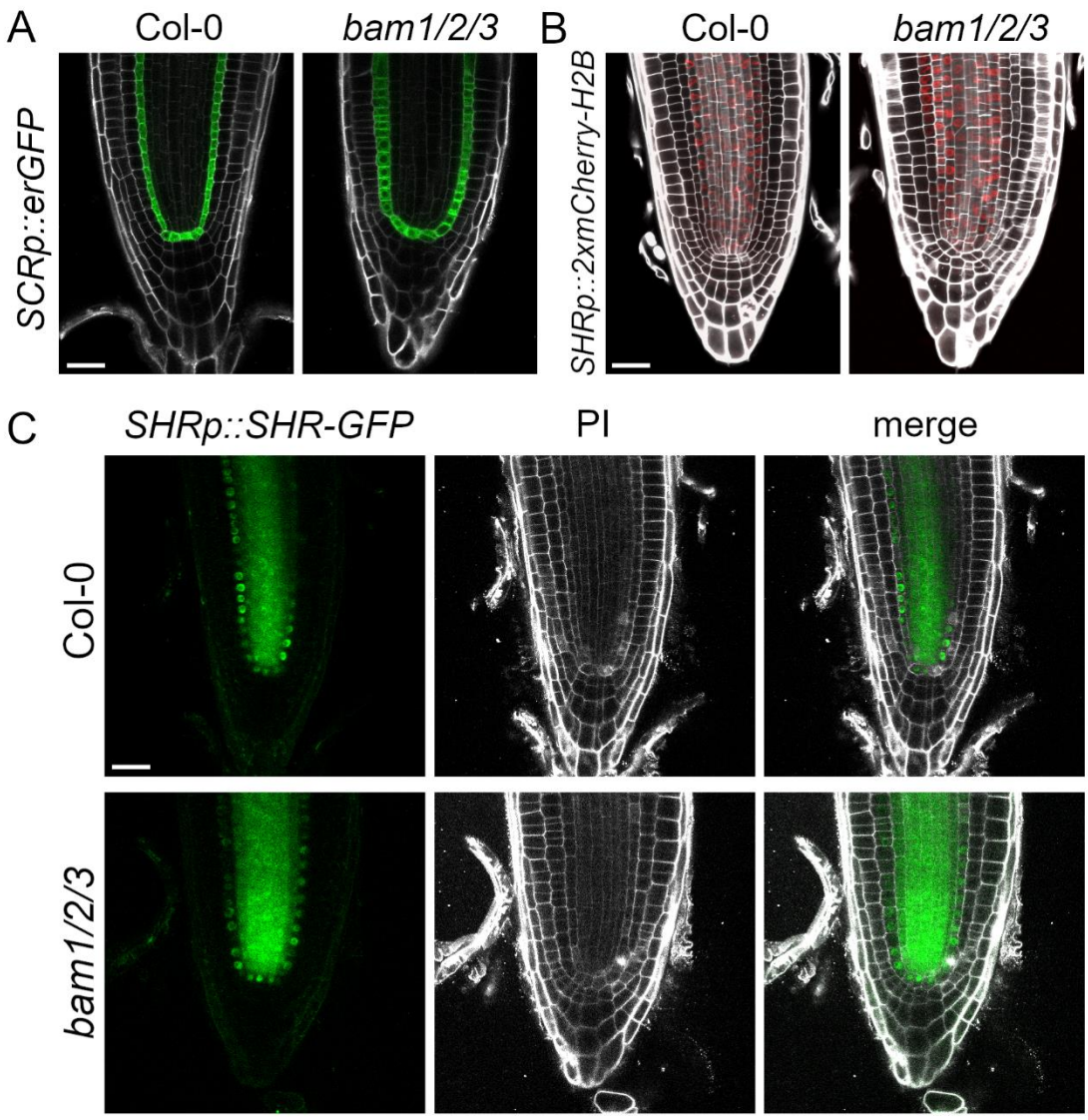

Fig. S2. BAM1/2 do not regulate *SHR* or *SCR* expression or protein dynamics. (A-B) Expression patterns of *SCR* (A; 7 dag) and *SHR* (B; 4 dag) were unchanged in *bam1/2/3* roots when compared to Col-0 roots. (C) SHR-GFP was localized in the stele of Col-0 and *bam1/2/3* and the subsequent trafficking into the ground tissue and nuclear sequestration was observed. Scale bar = 25 µm (A-C).

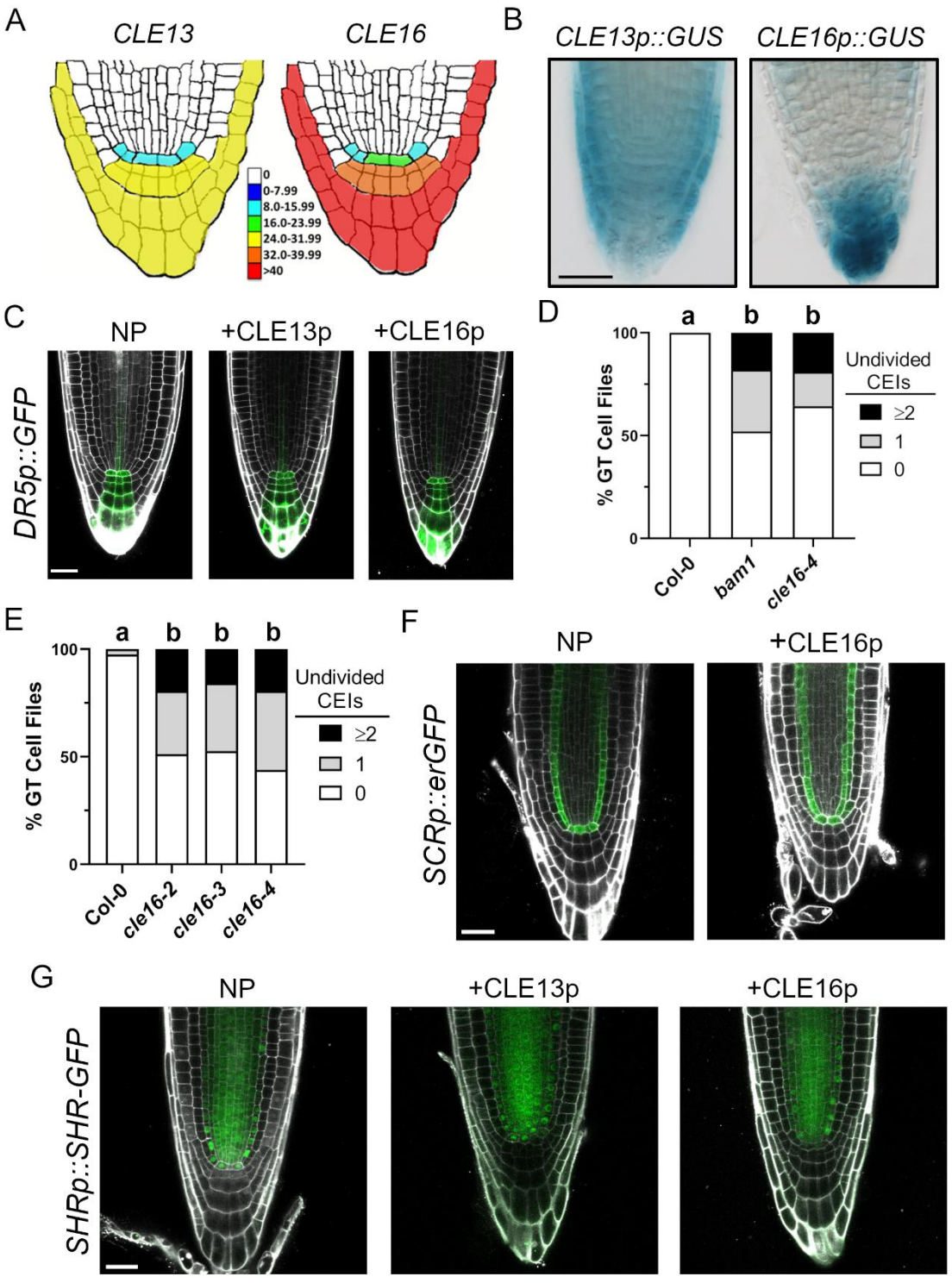

Fig. S3. Root enriched CLE genes regulate formative cell divisions. (A) Expression profiles of *CLE13* and *CLE16* by cell-type specific RNA-seq (1) (heatmap represents relative transcript abundance) and (B) *promoter::GUS* fusions in the root meristem. *CLE13* is largely expressed

throughout the stem cell niche, with *CLE16* expression is most prominent in columella and lateral root cap cells. (C) The expression pattern of *DR5p::GFP* does not change in response to peptide treatment in plants at 3 dag. (D) Undivided CEI quantification in the GT cell files of *bam1* and *cle16-4* (n= 32, Col-0; n= 50, *bam1*; n= 42, *cle16-4*) show similar division defects. (E) Three independent mutant alleles of *CLE16* display congruent phenotypes (n= 40, Col-0; n= 41, *cle16-2*; n= 38, *cle16-3*; n= 41, *cle16-4*). Distributions were compared using a Kruskal-Wallis nonparametric test. (F-G) Confocal imaging of roots expressing *SCRp::erGFP* (F) and *SHRp::SHR-GFP* (G) did not show altered patterns when treated with peptides. Scale bar = 25 µm (B, C, F, and G).

Table S1. Locus ID, sequence, and *CYCLIND6;1p::GFP* response of synthesized root expressed CLE peptides.

CLEp	Locus ID	Sequence	# AA	Upregulation of *CYCD6* observed?
CLE1p	At1g73165	RLSPGGPDPRHH	12	No
CLE2p	At4g18510	RLSPGGPDPQHH	12	No
CLE12p	At1g68795	RRVPSGPNPLHH	12	Yes
CLE13p	At1g73965	RLVPSGPNPLHH	12	Yes
CLE14p	At1g63245	RLVPKGPNPLHN	12	n.d.
CLE16p	At2g01505	RLVHTGPNPLHN	12	Yes

[n.d.] not determined

Table S2. Locus ID and allele information for genotypes used in the study.

Genotype	Locus ID	Mutant allele
BAM1	At5g65700	*bam1-4*, SALK_107290
BAM2	At3g49670	*bam2-4*, SAIL_1053_E09
BAM3	At4g20270	*bam3-2*, SALK_044433
		SALK_004121
SHR	At4g37650	*shr-2*, CS2972
SCR	At3g54220	*scr-3*, CS3997
CLE16	At2g01505	*cle16-2*
		cle16-3
		cle16-4

Table S3. Primer sequences used for cloning and genotyping.

Genotype		Primer sequence (5'-3')	Notes
BAM1	F	ccggtactctttccccagatgtttctcatttacgtc	
	R	cttattggaagagagatcgacgagatttagtttacc	
bam1-4	F	cttattggaagagagatcgacgagatttagtttacc	
	R	attttgccgatttcggaac	T-DNA LB primer (LB1.1a)
BAM2	F	tatggttcgctttggtattg	
	R	gttagctcgttaccggaaacc	
bam2-4	F	gaagtccagctgccagaaac	detects BASTA^r
	R	gcaccatcgtcaaccactac	
BAM3	F	ggtgaagataacacaaccccttagccgcttccaacg	
	R	ccggtactctttccccagatgtttctcatttacgtc	
bam3-2/ SALK_004121	F	ccggtactctttccccagatgtttctcatttacgtc	
	R	attttgccgatttcggaac	T-DNA LB primer (LB1.1a)
SHR/shr-2	F	actcctccgtccttcgactt	*shr-2* (large indel)
	R	tctgtggctgcagctgttac	
SCR/*scr-3*	F	tcacgggacttggtacttcc	CAPs; MaeII site is introduced in *scr-3*
	R	cttctcgatggtcctccaac	
CLE16/*cle16-2*	F	gaatccaaaacctgctctgc	MspI site at +59 is altered in *cle16-2* (+C)
	R	cgaaggagcagtcaacacct	
CLE16/*cle16-3*	F	gaatccaaaacctgctctgc	MspI site at +59 is altered in *cle16-3* (-G)
	R	cgaaggagcagtcaacacct	
CLE16/*cle16-4*	F	caaatcaaacagccatggaagcttgttccagaaccaga	dCAPs; BseL1 site at +45 is altered in *cle16-4* (+A)
	R	cttggagagagaccagacac	
CLE16p::CLE16	F	GGGGACAAGTTTGTACAAAAAAGCAG GCTTCACCtatgcacttaaagtgtggtaacactg	attB1-*CLE16* 5'promoter
	R	GGGGACCACTTTGTACAAGAAAGCTG GGTGgatcttcgaagaaatccatgcattcg	attB2-*CLE16* 3' UTR

SI References

1. Clark, N., *et al.* Stem-cell-ubiquitous genes spatiotemporally coordinate division through regulation of stem-cell-specific gene networks. Nat. Commun. **10**, 5574 (2019).