

Supplemental Figure 1. RopGEF7 expression pattern analysis

(A) RopGEF7-GUS expression in 5-d-old seedlings, GUS staining was for 16 h in(A)-(C).

**(B)** GUS activity was detected in meristemnoids and guard cells of 5-d-old seedlings, arrowhead indicates meristemnoid.

(C) GUS activity was detected in the style of the flowers.

Scale bars for (A) and (C), 200 µm. Scale bar for (B), 20 µm.



Supplemental Figure 2. Early embryo phenotypes are observed in the *RopGEF7RNAi* lines

(A) Relative transcript level of *RopGEF7* detected by qRT-PCR in embryos of four *RPS5A-RopGEF7RNAi* transgenic lines.

(**B**) Relative transcript level of *RopGEF1* was not affected in embryos of the *RPS5A-RopGEF7RNAi*-L6 line.

(C) - (F) Wild-type embryos at 8-cells (C), 16-cells (D), 32-cells (E), and globular stage (F), respectively.

(G)-(K) Defects in *RPS5A-RopGEF7RNAi* embryos at corresponding stages as compared to wild-type. Figures at right bottom indicate the number of embryo cells. Scale bars for (C)- (K), 20  $\mu$ m.

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Supplemental Figure 3. Analysis of RopGEF7 RNAi transgenic lines

(A) Identification of *35S-RopGEF7 RNAi* transgenic lines L4-1, L4-7, L4-8, L4-10, L4-12 by qRT-PCR analysis. qRT-PCR analysis revealed four lines with various levels of reduced *RopGEF7* transcripts compared with the wild-type.

(B)-(C) 5-d-old *RopGEF7* down-regulated lines show reduced root length (B) and smaller root meristem size compared with the wild type (C). Data presented are average and SD (n=40 to 60).

(**D**)-(**E**) Comparison of embryo phenotype from wild-type (**D**) and 35S-RopGEF7RNAi (**E**)-L1-9 line at heart stage. Bracketed area is the basal cell region.

Scale bars for (D)- (E), 20  $\mu$ m.



Supplemental Figure 4 Analysis of  $RopGEF7 \triangle C$  overexpressing transgenic plants

(A) Cotyledon epidermal cells of 7-d-old wild-type seedlings.

(**B**)-(**D**) Analysis of cell morphology in *RPS5A-RopGEF7* $\triangle C$  root-like structures, red dotted area in (**B**) indicate the root cell files that enlarged in insets, red dotted area in (**C**) indicates the root columella-like cells, (**D**) shows magnification of the tip region of the root-like structure in (**C**).

(E) Comparison of 8-d old seedlings in *RPS5A-RopGEF7* $\triangle C$  –L34 line and the wild-type.

(**F**)-(**G**) Analysis of root length (**F**) and lateral roots (**G**) in 8-d-old seedlings between *RPS5A-RopGEF7* $\triangle C$  –L34 line and the wild-type. Data presented are average and SD (n=40 to 60).

Scale bars for (A) to (D), 50  $\mu$ m.

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Supplemental Figure 5. Expression of *RopGEF7* in *plt1-4 plt2-2* double mutant

(A) Relative expression levels of *RopGEF7* detected by q-RT-PCR in 7-d-old seedlings of *plt1-4 plt2-2* compared with the wild-type.

(**B**)-(**E**) *RopGEF7-GUS* expression in embryos and 6-d-old seedlings of wild-type (**B** and **D**) and *plt1-4 plt2-2* (**C** and **E**), GUS staining was for 16 h in embryos, 6 h in seedlings.

Scale bars for (**B**) to (**E**), 20  $\mu$ m.

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Vector	GEF7promoter-F	5'-AAGCTTCTTCATTCATCCATCCCACCT -3'
constructions	GEF7promoter-R	5'-TCTAGACGTGCTTGAATCGCATTACAT -3'
	GEF7RNAi-F	5'-AAGCTTCTTACAAGACATTCATCGGA-3'
	GEF7RNAi-R	5'-GTCGACATCTCAAAGTCAAAGCCCAT-3'
	YFP-GEF7-F	5'-GAATTCATGGATGGTTCGTCGGAAAAT-3'
	YFP-GEF7-R	5'-GTCGACGTCAAAGCCCATCTTTCCAA-3'
	YFP-GEF7∆C-F	5'-AAGCTTATGGATGGTTCGTCGGAAAA-3'
	YFP-GEF7∆C-R	5'-GTCGACTCATCTTGTAAGATCATCAACAAAGAG-3'
	YFP-AtRAC1-F	5'-CTGCAGTCCATTTCTGGTGGAGAAGG -3'
	YFP- AtRAC1-R	5'-GTCGACTTGTTCCAGAGTTCGTTGTGA -3'
	GEF7OX-F	5'-GAATTCTAATGCGATTCAAGCACGAG-3'
	GEF7OX-R	5'-GTCGACGTCAAAGCCCATCTTTCCAA-3'
	GEF7∆COX-F	5'-GTCGACATGGATGGTTCGTCGGAAAA-3'
	GEF7∆COX-R	5'-GAATTCTCATCTTGTAAGATCATCAACAAAGAG-3'
	cYFP-GEF7-F	5'-GAATTCCATGGATGGTTCGTCGGAAAATT-3'
	cYFP-GEF7-R	5'-GTCGACGTCAAAGCCCATCTTTCCAA-3'
	cYFP-GEF7∆C-F	5'-GAATTCATGGATGGTTCGTCGGAAAA-3'
	cYFP-GEF7∆C-R	5'-GAATTCTCATCTTGTAAGATCATCAACAAAGAG-3'
	nYFP- AtRAC1-F	5'-GAATTCAGGAAGAAGAAGAAGAAATGAGCG-3'
	nYFP-AtRAC1-R	5'-GTCGACTCACAACGAACTCTGGAACAAT -3'
	RPS5A-F	5'- AGCAGCAGGAGATCTATCAGTGCA -3'
	RPS5A-R	5'- GGCTGTGGTGAGAGAAACAGAGC -3'
In situ	PLT1insitu-F	5'-ACGAAAACCAATCCAACCAC -3'
hybridization	PLT1insitu-R	5'-CCTAGACTGGCCTTCCCTTC -3'
RT-PCR	GEF7-F	5'-ATTCAACATTGTTGCACGCAT -3'
analysis of	GEF7-R	5'-CGGCTCGATCTTTCTAAAGGA-3'
RopGEF7	ACTIN2-F	5'-ATGGCTGAGGCTGATGATATTCAAC-3'
expression	ACTIN2-R	5'-TACAAGGAGAGAACAGCTTGGATG-3'
RT-qPCR	GEF7qPCR-F	5'-CTGTACGGAGGATTTCACGGC-3'
analysis	GEF7qPCR-R	5'-CTTCTCCAAGCAGCAGTTTCGA-3'
	GEF1qPCR-F	5'- CGGCGGCAAAGATGTGGTC-3'
	GEF1qPCR-R	5'-GATTGGTGATGGCGTTGGAGAT-3'
	ACTIN7qPCR-F	5'-AGCGATGGCTGGAACAGAAC-3'
	ACTIN7qPCR-R	5'-CCTTCGTCTTGATCTTGCGG-3'

## Supplemental Table 1: Primers used in the study

Genotype	Stage	Defects( def./t	Percentage(%)
		ot.)	
WT(Col.)	Before Heart <sup>1</sup>	0/248	0
	Heart	0/127	0
RPS5A-RopGEF7RNAi-L6	Before Heart <sup>1</sup>	16/332	4.8%
	Heart	30/156	$19.2\%^{2}$

	Supplemental Table 2	Frequencies of embryo d	lefects in RPS5A-RopGEF	7RNAi plants
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<sup>1</sup>Embryos were analyzed at proembryo, globular and triangular stages.

<sup>2</sup>Note that the frequency of RNAi heart embryos (in L6 line) with the basal defects is comparable to that of  $plt1^{-/-}plt2^{+/-}plt3^{-/-}bbm3^{-/-}$  mutants (Galinha et al., 2007).