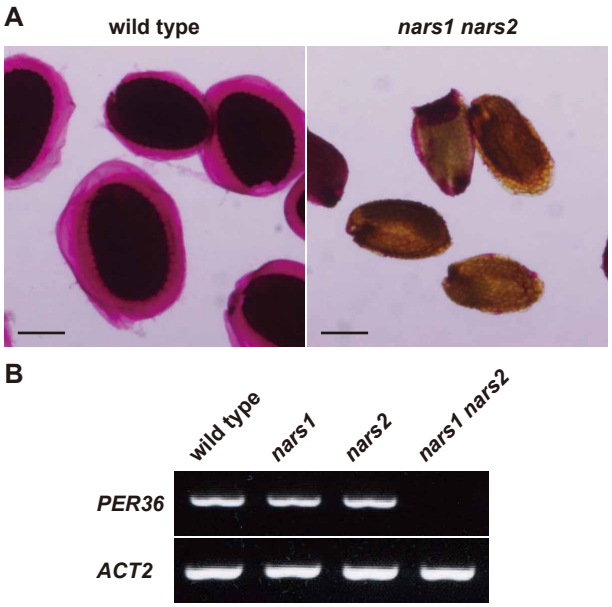
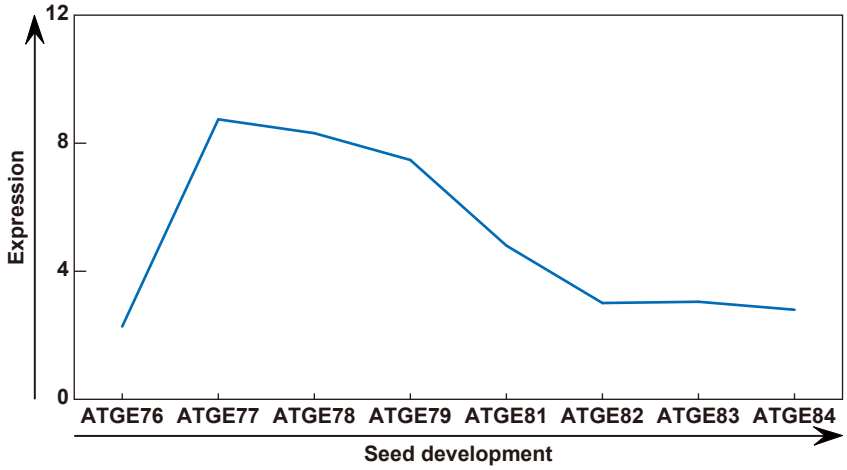
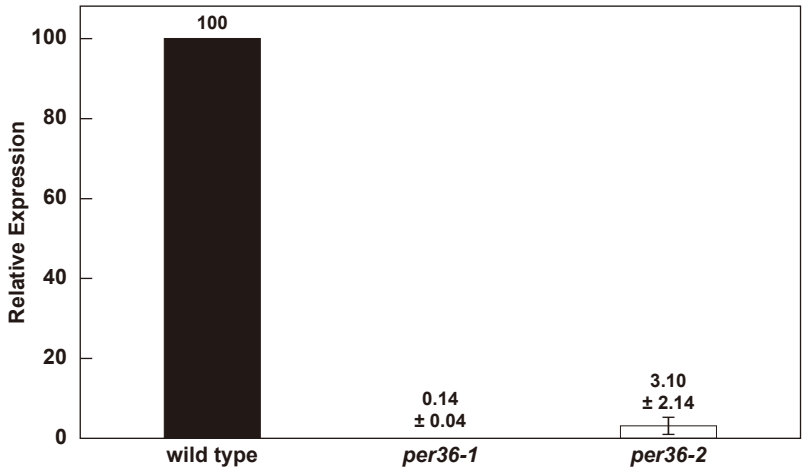


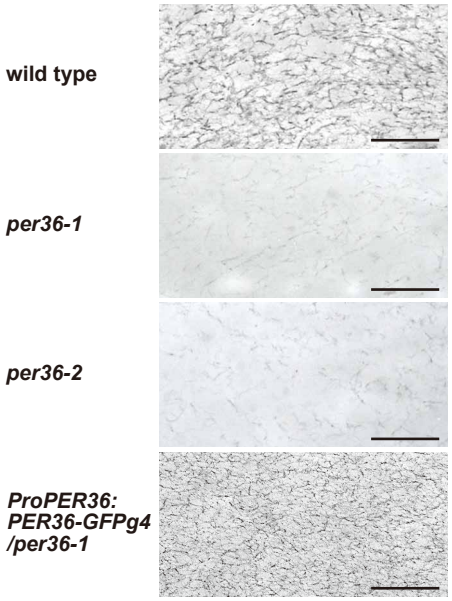
Supplemental Figure 1. NAC Transcription Factors NARS1 and NARS2 Regulate *PER36* Expression.
(A) Ruthenium red staining of dry seeds of wild type and the *nars1 nars2* double mutant. Mucilage extrusion was induced by shaking seeds in ruthenium red solution for 2 h. No mucilage extrusion was observed in the *nars1 nars2* double mutant, whereas wild-type seeds were surrounded by a large amount of mucilage. Bars = 200 μm.
(B) Gene expression analysis by RT-PCR showed that *PER36* was remarkably down-regulated in the *nars1 nars2* double mutant. RNAs were extracted from siliques containing developing seeds at the torpedo stage. RT-PCR with 35 cycles was performed. *ACT2* was used as an internal control of gene expression.



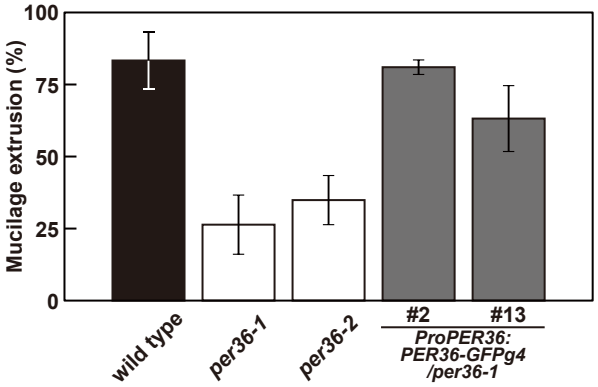
Supplemental Figure 2. Analysis of *PER36* Gene Expression during Early Seed Development.
The average expression values (log2 scale) of AtGenExpress (Schmid et al., 2005) are shown. Each ATGE corresponds to the following embryo stages: ATGE76, mid-globular to early heart embryos; ATGE77, early to late heart embryos; ATGE78, late heart to mid-torpedo embryos; ATGE79, mid- to late torpedo embryos; ATGE81, late torpedo to early walking-stick embryos; ATGE82, walking-stick to early curled cotyledons embryos; ATGE83, curled cotyledons to early green cotyledons embryos; ATGE84, green cotyledons embryos.



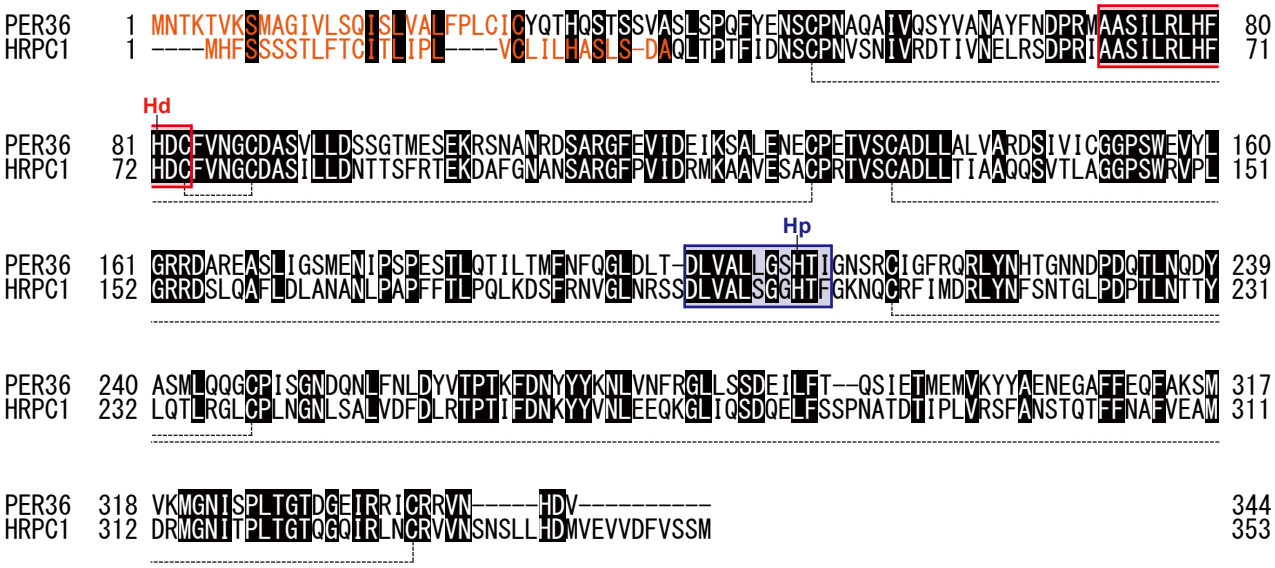
Supplemental Figure 3. Quantitative RT-PCR Showing the Expression Levels of *PER36* mRNA in Developing *per36-1* and *per36-2* Seeds at the Torpedo-Shaped Embryo Stage. *ACT2* was used as a standard to normalize the values. The mean values ± SD are calculated from three-independent biological replicates.



Supplemental Figure 4. Transmission Electron Micrographs Showing Filamentous Structures in the Mucilage of the Outermost Integument Cells.
Developing seeds at 9 DAP of the wild type, two *per36* mutant alleles and *ProPER36:PER36-GFPg4/per36-1* plants were used. Note that the impaired development of the structures in each *per36* mutant allele was rescued by expressing PER36-GFPg4. Bars = 500 nm.



Supplemental Figure 5. Comparison of Mucilage Extrusion in Dry Seeds of Wild Type, Two *per36* Mutant Alleles, and Two Independent Lines of *ProPER36:PER36-GFPg4/per36-1*.
The dry seeds were incubated in ruthenium red solution without shaking for 1 h. The seed staining patterns assessed as normal mucilage extrusion and partial mucilage extrusion were quantified together. Error bars indicate ±SD of three biological replicates.



Supplemental Figure 6. Comparison of the Amino Acid Sequences between PER36 and HORSERADISH PEROXIDASE C1 (HRPC1).
Closed boxes indicate identical amino acids between PER36 and HRPC1. Conserved amino acid sequences are marked as follows: a catalytic and distal heme-binding domain is indicated by a red box; a proximal heme-binding domain is indicated by a blue box; four disulfide bridges are indicated by dotted lines. Hd and Hp are distal and proximal histidine, respectively. The putative signal peptide (orange characters) was predicted by SignalIP 4.1 Server with a D-cutoff value of 0.34 (Petersen et al., 2011).

Supplemenatal References

Petersen, T.N., Brunak, S., von Heijne, G., and Nielsen, H. (2011). SignalP 4.0: discriminating signal peptides from transmembrane regions. Nat. Methods **8:** 785-786.

Supplemental Table 1. Monosaccharide Composition of Mucilage in Dry Seeds of the Wild Type and Two *per36* Mutant Alleles.

Sugar	Na_2CO_3 fraction			KOH fraction		
	wild type	*per36-1*	*per36-2*	wild type	*per36-1*	*per36-2*
Fuc	0.03 ± 0.01	0.04 ± 0.00	0.03 ± 0.01	0.21 ± 0.01	0.20 ± 0.01	0.22 ± 0.00
Ara	0.34 ± 0.05	0.32 ± 0.01	0.29 ± 0.04	6.19 ± 0.19	6.39 ± 0.23	6.36 ± 0.39
Rha	12.31 ± 0.71	12.56 ± 0.38	11.66 ± 0.63	10.36 ± 1.84	14.21 ± 1.38	15.20 ± 0.47
Gal	0.88 ± 0.08	0.78 ± 0.02	0.77 ± 0.04	5.38 ± 0.29	5.40 ± 0.28	5.69 ± 0.05
Glc	0.33 ± 0.05	0.29 ± 0.01	0.27 ± 0.00	5.37 ± 0.22	5.18 ± 0.20	5.44 ± 0.09
Xyl	1.21 ± 0.07	1.14 ± 0.04	1.08 ± 0.07	3.01 ± 0.31	2.93 ± 0.26	3.37 ± 0.07
Man	0.27 ± 0.02	0.25 ± 0.01	0.24 ± 0.01	2.88 ± 0.15	2.88 ± 0.22	3.04 ± 0.01
GalA	12.73 ± 0.45	12.02 ± 0.25	11.90 ± 0.28	10.49 ± 1.25	13.15 ± 0.88	13.58 ± 0.18

Monosaccharide quantities in the Na_2CO_3 and KOH fractions of the mucilage were determined by HPAEC. Details are in Materials and Methods. The mean values ± SE (μg/mg seed) are calculated from three-independent biological replicates (student's *t*-test between wild type and *per36* mutants, $P < 0.05$).

Supplemental Table 2. DNA Constructs Used in This Study

Plasmid name	Purpose	Plasmid backbone	Insert DNA or Entry clone	Primers	Cloning method
pDONR P4-P1R/ProPER36	Entry clone	pDONR P4-R1R	PER36 promoter (the 5'-upstream region of *PER36* including 2,635 bp from the site of translational initiation)	ProAt3g50990B4_F2 ProAt3g50990B1_R1 HybridGW-attB4_F HybridGW-attB1_R	Gateway BP
pENTR/sGFPΔstop	Entry clone	pENTR/D-TOPO	sGFP CDS except for the stop codon	sGFP-TOPO_F sGFP-TOPO_R1	TOPO cloning
pENTR/PER36GFPg4	Entry clone	pENTR/D-TOPO	PER36 CDS conjugated with GFPg4 CDS	At3g50990TOPO_F1 At3g50990_R1 At3g50990sGFP_F1 sGFP-G4_R2	TOPO cloning
R4pGWB533/ProPER36:sGFP-GUS	Plant transformation	R4pGWB533	pDONR P4-P1R/ProPER36 pENTR/sGFPΔstop	- -	Gateway LR
R4pGWB501/ProPER36:PER36GFPg4	Plant transformation	R4pGWB501	pDONR P4-P1R/ProPER36 pENTR/PER36GFPg4	- -	Gateway LR
pH2GW7/Pro35S:PER36GFPg4	Plant transformation	pH2GW7	pENTR/PER36GFPg4	-	Gateway LR

Supplemental Table 3. Primers Set Used in This Study

Primer name	Sequence	Purpose
ProAt3g50990B4_F2	5'-gtatagaaaagttgGGCCCATATAAGTTAA-3'	For amplifying 5' upstream region of PER36 gene with the adaptor sequence of BP reaction
ProAt3g50990B1_R1	5'-tttgtacaaacttgTTGGACTCTCAGCGA-3'	For amplifying 5' upstream region of PER36 gene with the adaptor sequence of BP reaction
HybridGW-attB4_F	5'-GGGGACAACTTTGTATAGAAAAGTTG-3'	For conjugating the adaptor sequence of BP reaction
HybridGW-attB1_R	5'-GGGGACTGCTTTTTTGTACAAACTTG-3'	For conjugating the adaptor sequence of BP reaction
sGFP-TOPO_F	5'-ccacATGGTGAGCAAGGGCGA-3'	For amplifying CDS sequence of sGFP gene with the 4 bp sequence (cacc)
sGFP-TOPO_R1	5'-GAGATCTCCCTTGTACAGCTCGT-3'	For amplifying CDS sequence of sGFP gene without the stop codon
At3g50990TOPO_F1	5'-caccATGAATACAAAAACGGTGAAG-3'	For amplifying CDS sequence of PER36 gene with the 4 bp sequence (cacc)
At3g50990_R1	5'-AACATCATGGTTAACCCTCCGGCAT-3'	For amplifying CDS sequence of PER36 gene without the stop codon
At3g50990sGFP_F1	5'-AGGGTTAACCATGATGTTATGGTGAGCAAG-3'	For amplifying and conjugating the full-length coding regions of PER36 and sGFP
sGFP-G4_R2	5'-tcatcctcctcctccGAGATCTCCCTTGTA-3'	For amplifying CDS sequence of sGFP gene with the 4 residues of glycine
PER36RT_F1	5'-CTCACTTGTTGCGCTGTTTCCT-3'	For amplifying endogenous transcript of PER36 by RT-PCR
PER36RT_R1	5'-TCCGGATCTCACCATCCGTC-3'	For amplifying endogenous transcript of PER36 by RT-PCR
At3g15510RT_F1	5'-CCCTACCGACGAAGAGCTTGTTG-3'	For amplifying endogenous transcript of NARS1 by RT-PCR
At3g15510RT_R1	5'-CAATGGTGGAGTCTCTTCCATCATG-3'	For amplifying endogenous transcript of NARS1 by RT-PCR
AtPDF1_F	5'-ACTCCGGTTGTTGTGACTCC-3'	For amplifying endogenous transcript of PDF1 by RT-PCR
AtPDF1_R	5'-GTGCCTTCACGGTAGAGAGC-3'	For amplifying endogenous transcript of PDF1 by RT-PCR
dVPE_F	5'-ATGTCTAGTCCTCTTGGTCACTTTC-3'	For amplifying endogenous transcript of deltaVPE by RT-PCR
dVPE_R	5'-CAAGCTTGTTCAATGGCTGA-3'	For amplifying endogenous transcript of deltaVPE by RT-PCR
Actin2-F	5'-AGAGATTCAGATGCCCAGAAGTCTTGTTCC-3'	For amplifying endogenous transcript of ACT2 by RT-PCR
Actin2-R	5'-AACGATTCCTGGACCTGCCTCATCATACTC-3'	For amplifying endogenous transcript of ACT2 by RT-PCR