Supplemental data

Supplemental Figure S1



Supplemental Figure S1. Turgor-dependent translocation of VIP1-GFP in a hypocotyl and a leaf. Hypocotyls and leaves of VIP1-GFPox #2 plants were observed by epifluorescent microscopy at the indicated time points after being submerged in 20 mM Tris-HCl, pH 6.8 (rehydration). Scale bars = $100 \mu m$.



Supplemental Figure S2. Turgor-dependent translocation of VIP1-GFP is not affected by ABA or gibberellin A3. Roots of VIP1-GFPox #2 plants were observed by epifluorescent microscopy at the indicated time points after being submerged in 20 mM Tris-HCl, pH 6.8, containing 100 μ M ABA (+ ABA) or 0.1 mg/mL gibberellin A3 (+ GA3). Scale bars = 100 μ m.

TCG-TCTCTCTCAAATGAGCTGTCTCTCTAATAAGAGTTTCTTCTGTATAGGAAAGGAA---AGGTCTCT<u>CTCTCTC</u> TCACTCTCTCCAAATTAGCTGGCTCC--AAAAAAGGTTCCTTTTAGGAAACTCCCTTCTCCAATTTGTTTCCTCAT

CCATTACATTAAAACTCCAAAAAATTCATTTTGTTTTCTTTTAGAGTTCACAAGTTCT<u>TCGTTGTTCAGCTACTCC</u> CCATTAGAGAGAGAA<u>ACTCACAAAACATACTTCGAATTCCCA</u>TT---GTTTAAAAGACGAAGATA------------

Supplemental Figure S3. Alignment of DNA sequences of the *CYP707A1* promoter (upper rows) and the *CYP707A3* promoter (lower rows). Approximately 350-bp sequences upstream of the start codon were aligned. Gray boxes indicate DNA residues shared between both promoters. Black boxes indicate putative transcriptional initiation positions. G-box (CACGTG) is highlighted by rectangles. Primers used for making probes for the gel shift assays are shown by underlines (solid lines: forward primers; broken lines: reverse primers).



Supplemental Figure S4. Relative growths of VIP1-GFPox plants under mannitol stress conditions. In each genotype, the fresh weight under mannitol stress conditions was divided by the fresh weight under the control (mannitol-free) condition and the resultant value is shown as relative growth (thus the value is 100% for the control condition in all the genotypes). The values shown in Fig. 9B were used as the fresh weights for calculation.



Supplemental Figure S5. Responses of VIP1-GFPox plants to NaCl stress. Images of

2-week-old plants grown on media with 0 (Control) and 125 mM NaCl.



Supplemental Figure S6. Expression levels of dehydration-responsive genes, *NCED3*, *RD29A* and a rehydration-responsive gene, *PDH1*, under mannitol stress. Plants were grown for 3 weeks on media with 0 (Control) and 200 mM mannitol and used for RNA isolation and cDNA synthesis. Relative expression levels were calculated by the comparative C_T method using *UBQ5* as an internal control gene and the sample of wild type grown under the control condition as a reference sample. Experiments were performed in triplicate. Values are means \pm SE.



Supplemental Figure S7. Induction of *NCED3*, *RD29A*, *RAB18* and *RD29B* by ABA. Two-week-old wild-type seedlings were incubated for 90 minutes at room temperature in 20 mM Tris-HCl, pH 6.8, with 0 (Mock) or 100 μ M ABA (+ ABA), and used for RNA isolation and cDNA synthesis. Relative expression levels were calculated by the comparative C_T method using *UBQ5* as an internal control gene and the mock-treatment sample as a reference sample. Experiments were performed in triplicate. Values are means ± SE.

Gene		Primer sequence $(5' > 3')$
VIP1-GFP	Fw	CGGCGTCGTTTAATATCGAATC
	Rv	GTAGGTGGCATCGCCCTCGC
VIP1 (ORF)	Fw	CGGCGTCGTTTAATATCGAATC
	Rv	CAGCCTCTCTTGGTGAAATCC
VIP1 (UTR)	Fw	CGGCGTCGTTTAATATCGAATC
	Rv	GACCGATGTCGCCATGAACATA
VIP1	Fw	AGCGCTGCGGGGATGAACTGA
(for qRT-PCR)	Rv	GGAGCGATGGCTGCCCGTTT
CYP707A1	Fw-1 ^a	GCCGCTAGAGACACGACGGC
	Fw-2 ^a	TGGAACCCACTCGTGTCCTGGA
	Rv	CCCGTCGCTCGCTCCAACAA
<i>CYP707A3</i>	Fw-1 ^a	CGCAAGAGACACGACGGCGA
	Fw-2 ^a	TCGAAGTTGCGCCGAAACCGA
	Rv	GGCCCTACGATTGACCATCTGTACT
MYB44	Fw	CACCACTTCGAGCCGCCACA
	Rv	CCTCCGACGAATCCGCCACC
NCED3	Fw	TGGCGAAGCAGGTCGTGTGA
	Rv	ACCAGCTGAGCTCGAACGGC
RD29A	Fw	GCCGGAATCTGACGGCGGTT
	Rv	CCCGTCGGCACATCCTTGTCG
RAB18	Fw	TCCAGCTCTAGCTCGGAGGATGA
	Rv	GGATCCCATGCCGCCCATCG
RD29B	Fw	AACCGAAGTCGCCACGGTCC
	Rv	CTCCGCCACTGCCTCCCAACT
PDH1	Fw	CCCTGGGTTGCAAGTCGCCA
	Rv	CCGGTGGCCATCATTCCCCG
UBQ5	Fw	GACGCTTCATCTCGTCC
	Rv	CCACAGGTTGCGTTAG

Supplemental Table S1. Primer pairs used for RT-PCR analyses

^aFw-1 primers were used for semi-quantitative RT-PCR and Fw-2 for quantitative RT-PCR.