## SUPPLEMENTAL DATA

**Supplemental Figure S1: Expression analysis of additional genes with shoot-specific response to hypoxia.** Expression of selected hypoxia-responsive genes determined after treatment of seedlings with hypoxia for 2 and 8 hours in either light or darkness. Roots (R) and shoots (S) were harvested separately. One representative experiment out of three independent replicates is shown. The photo shows the RT-PCR bands separated by agarose gel electrophoresis and after staining with Serva DNA stain G.

	2 hours							8 hours					
	Light				Da	ark	Light			D	Dark		
	С		hypo		oxia		С		hypo		oxia		
	S	R	S	R	S	R	S	R	S	R	S	R	
Tubulin													
DGD1													
DGD2													
SQD1		-	-										
SQD2	in de la composition Recorder de la composition Recorder de la composition										an a		
G3Pp1			-						-				
SRG3													

Supplemental Figure S2: The gene expression changes are not due to  $CO_2$  deficiency. Expression of selected hypoxia-responsive genes determined after treatment of seedlings with hypoxia for 2 and 8 hours in light, supplemented with  $CO_2$  through the N<sub>2</sub> gas flow (+ $CO_2$ ) or not (- $CO_2$ ). One representative experiment out of three independent replicates is shown. The photo shows the RT-PCR bands separated by agarose gel electrophoresis and after staining with Serva DNA stain G.

		2 hours		8 hours			
	control	hypo	oxia	control	hypo	oxia	
	+CO <sub>2</sub>	-CO <sub>2</sub>	+CO <sub>2</sub>	+CO <sub>2</sub>	-CO <sub>2</sub>	+CO <sub>2</sub>	
Tubulin							
LBD41				-			
MGD2							
MGD3			and the Landour				
At5g20790							
PAP17			-	even same			

Supplemental Figure S3: Lipid composition of membranes after submergence. Molecular species composition [mol%] of different membrane lipids in leaves kept under submergence or control conditions for 3 and 7 days. Data are means  $\pm$  SD for five replicates. Asterisks mark significant differences between control and submerged conditions for each lipid (P<0.05, Tukey HSD test).



**Supplemental Figure S4: Overlap of gene expression with other stresses.** Heat map of the 51 genes that are induced shoot-specific during two hours of hypoxia. Data are signal-log ratios of different stresses or genotypes versus respective controls and were obtained through the Genevestigator website (https://www.genevestigator.com/gv/plant.jsp, Hruz et al., 2008). Red color represents a signal-log-ratio of >0, green represents a signal-log-ratio of <0.



**Supplemental Figure S5: Expression analysis of additional low-Pi-responsive genes under hypoxia and phosphate deficiency.** Expression of selected hypoxia-responsive genes determined after treatment of seedlings with hypoxia (H) for four hours or with phosphate deficiency (- Pi) for 48 hours in light. One out of two independent experiments with two individual samples is shown. The photo shows the RT-PCR bands separated by agarose gel electrophoresis and after staining with Serva DNA stain G.



**Supplemental Figure S6: Gene expression in different signaling mutants.** Heat map of 51 genes that are induced shoot-specific during two hours of hypoxia. Data are signal-log ratios of a comparison of stress treatment versus control or genotype versus wildtype. Hypoxia and phosphate deficiency data are from Mustroph et al. (2009) and Bustos et al. (2010), respectively. Mutant data were from the following sources: *prt6*, *ate1/2*, Gibbs et al. (2011); Rap2.12 OE, Licausi et al. (2011); *phr1*, *phl1*, Bustos et al. (2010). Yellow color represents a signal-log-ratio of >0, blue represents a signal-log-ratio of <0. The list of genes is presented in **Supplemental Table S1**.



**Supplemental Figure S7: Photoreceptors are not important for hypoxic gene induction under illumination.** Expression of selected genes determined after treatment of wildtype and photoreceptor mutant (*cry1/2*, *phot1/2*, *phyA/B*) seedlings with hypoxia for four hours in light. The experiment was performed two times and one representative experiment with two samples is shown. The photo shows the RT-PCR bands separated by agarose gel electrophoresis and after staining with Serva DNA stain G.

	W	ΥT	cry	cry1/2 phot1/2		phyA/B		
	control	hypoxia	control	hypoxia	control	hypoxia	control	hypoxia
Tubulin			annin - annada	Second Second				
LBD41								-
MGD2	Sector more					territari territari		
MGD3						-		
At5g20790			gerren annen					
PAP17	Network America	inerana ana	interest and				second and	

**Supplemental Figure S8: The effect of norflurazon-mediated inhibition of photosynthesis on the phosphate deficiency response.** Expression of selected genes determined after treatment of wildtype *phl1* and *phr1B* seedlings with phosphate deficiency for 48 hours in light. Before stress treatment, plants were placed on norflurazon- (NF) containing medium or control medium for two days to inhibit carotenoid biosynthesis. One experiment out of two independent replications is shown. The photo shows the RT-PCR bands separated by agarose gel electrophoresis and after staining with Serva DNA stain G.

	C	ol-0	p	hl1	phr1B		
	- NF	+ NF	- NF	+ NF	- NF	+ NF	
	+Pi -Pi	+Pi -Pi	+Pi -Pi	+Pi -Pi	+Pi -Pi	+Pi -Pi	
Tubulin						-	
MGD3							
MGD2			handinga kananga			Sectored Sectored	
At5g20790							
PAP17		e anderna andran	hand the second	-	<u> </u>		
PHR1							
PHL1	Married Access	• (					

Supplemental Figure S9: Metabolite levels under hypoxia and phosphate deficiency. Levels of adenylates, sugars, starch and selected phosphorylated metabolites in seedlings after treatment with control (C) or hypoxia (H) for two hours or with phosphate deficiency (- Pi) for 48 hours in light or darkness. For hypoxia, roots (R) and shoots (S) were harvested separately, for phosphate deficiency whole seedlings were harvested. Data are mean values +/- SD of 5 to 7 replicates. Values with different letters represent significant differences between four treatment conditions for each tissue type (P<0.05, Tukey HSD test). n.s., no significant changes.



**Supplemental Table S1: Expression data for the 51 genes of the heatmaps in Fig. 1 and 3.** Signal-log-ratios of stress treatment versus control or mutant versus wildtype are shown. Genes were selected based on the criteria described in the Result section. Numbers represent the position in the heatmaps. The sources of the data are as follows: Misson et al. (2005), Mustroph et al. (2009), Bustos et al. (2010), Gibbs et al. (2011), Licausi et al. (2011), Woo et al. (2012).

Supplemental Table S2: Selection of oxygen- or phosphate-deficiency response genes presented in Fig. 3C. Signal-log-ratios of stress treatment versus control and adjusted P-values are shown. Genes were selected based on the following criteria: SLR>1,  $P_{adj.}$ <0.01. The sources of the data are Mustroph et al. (2009) and Bustos et al. (2010).

Supplemental Table S3: Modification of photosynthesis pigments under submergence. Pigment composition [ $\mu$ mol or nmol/ g FW] in leaves kept under submergence or control conditions for 1, 3 and 7 days, and of seedlings treated with hypoxia for 2-8 or 24 hours. The 8-h-value represents a value that was harvested one hour after the end of the day. Data are means  $\pm$  SD for four biological replicates. Different letters show significant differences between control and submerged conditions for each pigment (P<0.05, Tukey HSD test).

Supplemental Table S4: Primers and Arabidopsis accession numbers used in this study.

## SUPPLEMENTAL REFERENCES

- Bustos R, Castrillo G, Linhares F, Puga MI, Rubio V, Pérez-Pérez J, Solano R, Leyva A,
  Paz-Ares J (2010) A central regulatory system largely controls transcriptional activation and repression responses to phosphate starvation in Arabidopsis. PLoS Genet 6: e1001102
- Gibbs DJ, Lee SC, Isa NM, Gramuglia S, Fukao T, Bassel GW, Correia CS, Corbineau F, Theodoulou FL, Bailey-Serres J, et al (2011) Homeostatic response to hypoxia is regulated by the N-end rule pathway in plants. Nature **479**: 415–418
- Hruz T, Laule O, Szabo G, Wessendorp F, Bleuler S, Oertle L, Widmayer P, Gruissem W, Zimmermann P (2008) Genevestigator v3: a reference expression database for the metaanalysis of transcriptomes. Adv Bioinformatics 2008: 420747

- Licausi F, Kosmacz M, Weits DA, Giuntoli B, Giorgi FM, Voesenek LACJ, Perata P, van Dongen JT (2011) Oxygen sensing in plants is mediated by an N-end rule pathway for protein destabilization. Nature 479: 419–422
- Misson J, Raghothama KG, Jain A, Jouhet J, Block MA, Bligny R, Ortet P, Creff A, Somerville S, Rolland N, et al (2005) A genome-wide transcriptional analysis using *Arabidopsis thaliana* Affymetrix gene chips determined plant responses to phosphate deprivation. Proc Natl Acad Sci USA 102: 11934–11939
- Mustroph A, Zanetti ME, Jang CJH, Holtan HE, Repetti PP, Galbraith DW, Girke T, Bailey-Serres J (2009) Profiling translatomes of discrete cell populations resolves altered cellular priorities during hypoxia in Arabidopsis. Proc Natl Acad Sci USA **106**: 18843– 18848
- Woo J, MacPherson CR, Liu J, Wang H, Kiba T, Hannah MA, Wang X-J, Bajic VB, Chua N-H (2012) The response and recovery of the *Arabidopsis thaliana* transcriptome to phosphate starvation. BMC Plant Biol 12: 62