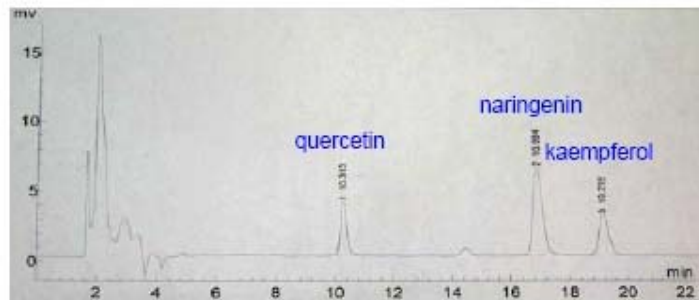


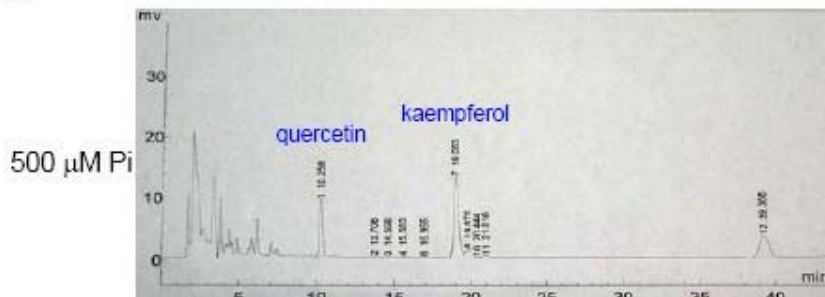
Accessory Publication

(a)

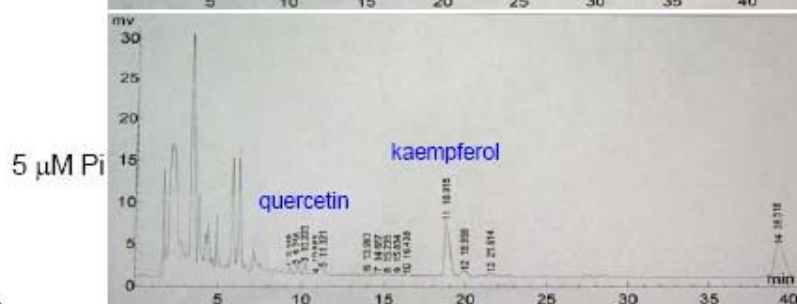


Flavonoid aglycone standards

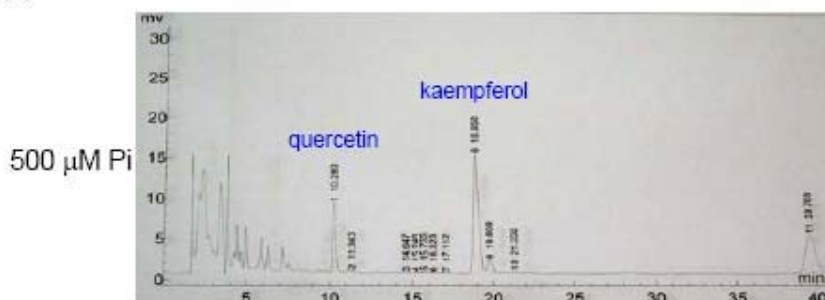
(b)



Root samples from wild type seedlings



(c)



Root samples from *pld2* seedlings

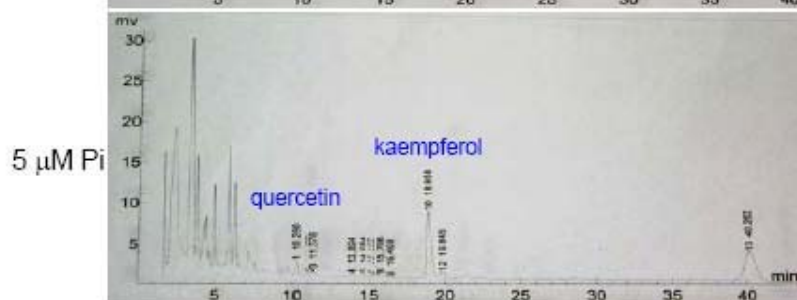


Fig. S1. Typical HPLC chromatograms of major aglycone flavonoids (quercetin, kaempferol) prepared from *Arabidopsis* seedling roots grown with high (500 μM) or low (5 μM) Pi concentrations. The peaks representing the major aglycones are labeled numerically according to their retention times. The identification of individual aglycone flavonoids was aided by both its retention time and specific absorption spectrum (not shown). (a) Commercial preparations of quercetin, naringenin and kaempferol. (b) Major aglycone flavonoids (quercetin, kaempferol) prepared from wild type *Arabidopsis* seedling roots grown with 500 (top panel) or 5 (lower panel) μM Pi. (c) Major aglycone flavonoids (quercetin, kaempferol) prepared from *pld ζ 2* seedling roots grown with 500 (top panel) or 5 (lower panel) μM Pi. The scale of the horizontal axis in (a) differs from that in (b) and (c). But this does not affect comparisons of the retention times for quercetin and kaempferol peaks.

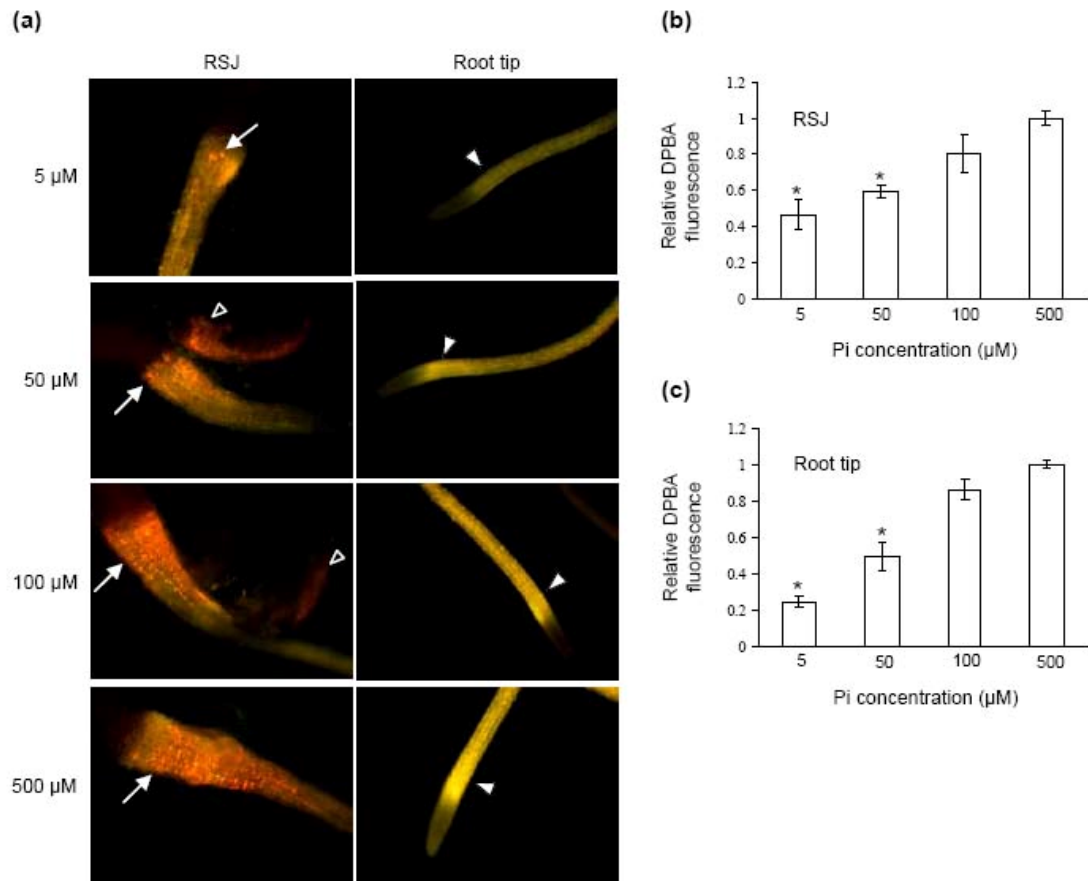


Fig. S2. Effects of Pi deprivation on the root flavonoid level of *Lerecotype* of *Arabidopsis* examined using DPBA staining. Four-day-old seedlings grown on the media with four Pi concentrations were treated with the flavonoid specific stain DPBA. (a) DPBA-elicited gold fluorescence (indicated by arrows) in the root-shoot junction (RSJ) and yellow fluorescence (marked by filled arrowheads) in the root tip regions. The seed coat structure (indicated by open arrowheads) is present in some preparations. The data displayed are representative of five sets of independent staining experiments. In each experiment, 10 (or more) different root samples were examined for each of the four Pi concentrations. (b) Quantitative comparisons of relative DPBA fluorescence levels in the RSJ region of *Ler* seedlings grown with four different Pi concentrations. (c) Quantitative comparisons of relative DPBA fluorescence levels in the root tip region of *Ler* seedlings cultured with four different Pi concentrations. In (b) and (c), means \pm S.D. were calculated using the measurements from 10 (or more) seedlings per Pi concentration per region (RSJ or root tip). The data set displayed is typical of three independent experiments. Asterisks indicate significant differences ($P \leq 0.05$) from the data determined for the roots grown with 500 μ M Pi.

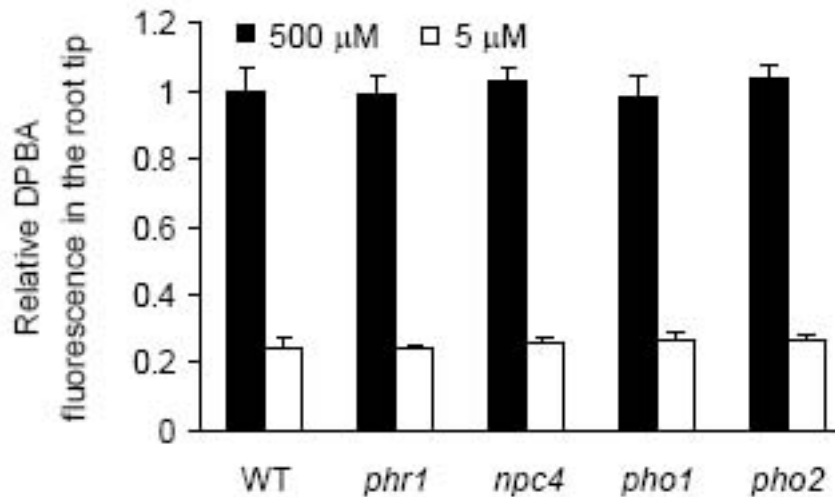


Fig. S3. Comparative analysis of the effects of low Pi treatment on the root flavonoid levels of wild type *Arabidopsis* (WT, Col-0 ecotype) and the knockout lines of PHR1 (*phr1*), NPC4 (*npc4*), PHO1 (*pho1*) or PHO2 (*pho2*). Four-day-old seedlings germinated on the media with high (500 μM , filled column) or low (5 μM , empty column) Pi concentrations were treated with the stain DPBA. The resulted flavonoid-specific fluorescence in the root-shoot junction (RSJ) and root tip regions was quantified. Means \pm s.d. were calculated using the measurements from 15 RSJ or roottip samples per genotype per Pi concentration. The data set shown is typical of three independent experiments. No significant differences were found among WT control, *phr1*, *npc4*, *pho1* and *pho2* in the degree of the reduction of root tip flavonoid level by low Pi. The five genotypes also showed a similar magnitude of reduction of flavonoid level in the RSJ region under 5 μM Pi (data not shown).

Table S1. Oligonucleotide primers used in the real-time PCR experiments of this study

Gene	Locus	Forward primer (5'-3')	Reverse primer (5'-3')
<i>CHS</i>	<i>At5g13930</i>	CGGTACTGTCCTCCGTATCG	CAAATGTCCGTCTATGGCACC
<i>CHI</i>	<i>At3g55120</i>	TCACCGGTGCGTTTGAGA	CGCCTCCGCCAACAATTT
<i>FLS</i>	<i>At5g08640</i>	TIAGGGTTAAAGCGTGATGCG	GGCGGAGGGAATATACTCTG
<i>F3H</i>	<i>At3g51240</i>	CGATACTAACTTGGTGGCGG	CGACGCATGCATTGGTAAGA
<i>F3'H</i>	<i>At5g07990</i>	CGGATATAATGGTTAAAGCC	AGCTCTCTGACGCGATGTGT
<i>DFR</i>	<i>At5g42800</i>	CTTATCACCGCGCTCTCTCC	AATGGTTGCATCATGAGAG
<i>Tubulin</i>	<i>At4g14960</i>	TGAGGTTGATGGTGCCTTG	TGTACTTTCCATGTCGCGGG