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Pi sensing and signaling: from prokaryotic to eukaryotic cells

WANJUN QI[1,2*], STEPHEN A BALDWIN[1], STEPHEN P MUENCH[1] and ALISON BAKER[2]

[1]*School of Biomedical Sciences, and Astbury Centre for Structural Molecular Biology, Faculty of Biological Sciences, University of Leeds, Leeds, LS2 9JT, UK*
[2]*School of Molecular and Cellular Biology, Centre for Plant Sciences and Astbury Centre for Structural Molecular Biology, Faculty of Biological Sciences, University of Leeds, Leeds, LS2 9JT, UK*
* Corresponding Author Email: bswq@leeds.ac.uk

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Abbreviations:

Pi: Phosphate;

Pst: Phosphate-specific ABC transport;

Pit: Phosphate inorganic transporter;

TF: Transcriptional Factor;

CDK: Cyclin Dependent Kinase;

CKI: Cyclin dependent Kinase Inhibitor;

IP7: inositol heptakisphosphate;

PSI: Pi Starvation Induced;

poly P: poly Phosphate;

SPX: Syg1-Pho81-Xpr;

Vtc: Vacuolar transporter chaperone;

P1BS: PHR1-Binding Site;

EXS: ERRD 1-XPR1-SYG1.

Abstract

Phosphorus is one of the most important macronutrients and is indispensable for all organisms as a critical structural component as well as participating in intracellular signaling and energy metabolism. Sensing and signaling of phosphate (Pi) has been extensively studied and is well understood in single-cellular organisms like bacteria (*Escherichia coli*) and *Saccharomyces cerevisiae*. In comparison, the mechanism of Pi regulation in plants is less well understood despite recent advances in this area. In most soils the available Pi, limits crop yield, therefore a clearer understanding of the molecular basis underlying Pi sensing and signaling is of great importance for the development of plants with improved Pi use efficiency. This mini-review compares some of the main Pi regulation pathways in prokaryotic and eukaryotic cells and identifies similarities and differences among different organisms, as well as providing some insight into future research.

Pi regulation in prokaryotic cells

In bacterial cells, Pi regulation typically involves Pi sensing and transport by a cytoplasmic membrane anchored protein complex as well as a two-component intracellular Pi signaling pathway, which modulates the expression of various genes that comprise the Pho regulon [1] (Fig. 1). In *E. coli*, sensing and transport of environmental Pi is primarily accomplished by a high-affinity **P**hosphate-**s**pecific ABC **t**ransport (Pst) system, which consists of a periplasmic Pi binding protein (PstS), a Pi channel formed by 2 membrane proteins (PstA and PstC), and a cytoplasmic dimeric ATPase (PstB) which provides energy for Pi transport across the membrane [2]. Under conditions where environmental Pi availability is above 4 μM, Pi binding to PstS inhibits the inner membrane Pi sensory histidine kinase PhoR in a Pi-uptake independent manner [1,3] subsequently abolishing the activation of downstream Pho-controlled gene expression. Although the exact mechanism of this inhibition is unclear, it is well established that a chaperone/Hsp70-like protein PhoU plays a critical role. It has been proposed that PhoU regulates the activity of PhoR by forming a complex between the PstB protein of the Pi transporter and the PAS (**P**er-**A**RNT-**S**im) domain of PhoR [3,4]. This hypothesis is also supported by a recent study from Gardner's group, which shows the binding of manganese and magnesium to dimeric PhoU via highly conserved residues which could be important for its membrane complex association [5]. However, the most recent study in *Caulobacter crescentus* suggested rather than negatively regulating the Pho regulon, PhoU modulates Pi uptake by the Pst transporter (Fig. 1) and prevents high intracellular Pi toxicity [6]. Further evidence is needed to confirm the precise function of PhoU and whether it plays diverse roles in different organisms.

Under Pi limited conditions, the repression on PhoR is released by disassociation from PhoU, thus allowing the auto-phosphorylation of this histidine kinase, and activation of its downstream response regulator PhoB [1] (Fig. 1). The transcription factor PhoB belongs to the OmpR/PhoB

family, and is characterized by an N-terminal receiver domain and a C-terminal DNA binding domain which contains a unique wHTH (**w**inged-**H**elix-**T**urn-**H**elix) domain [7]. A highly conserved Asp residue of PhoB is phosphorylated during low Pi stress and the phosphorylated PhoB subsequently activates the Pho regulon by binding to a specific Pho box sequence in the promoter region [3,7]. Computational predictions based on known Pho box sequences revealed 96 putative Pho regulated genes in the genome of *Sinorhizobium meliloti* and more than 100 in *E. coli*. Among these sequences are the genes which encode the Pst Pi sensing/transporting complex, integral membrane protein PhoR and transcription factor PhoB itself [8], indicating the existence of a positive feedback loop of bacterial Pi regulation.

The low-affinity **P**hosphate **i**norganic **t**ransporter (Pit) system also contributes to Pi uptake under Pi sufficient conditions (> 4 μM) (Fig. 1). However, this system primarily transports metal-phosphate complexes and its expression is largely controlled by the external Zn^{2+} level [9]. A recent study also shows the Pit system plays essential roles in *E. coli* copper tolerance by mediating Cu^{2+}-phosphate efflux from the cell [10].

Pi regulation in Yeast (*Saccharomyces cerevisiae*)

The topic of Pi regulation in the yeast *S.cerevisiae* has been reviewed in detail in a recent publication [11], to which the reader is referred for a more extensive discussion than is possible within the constraints of this short review . In this organism external Pi availability is sensed by plasma membrane localized proteins Pho84 and Pho87. Pho84 is both a phosphate transporter and a phosphate sensor which can couple growth to Pi availability via activation of the protein kinase A pathway. Thus it is often termed a 'transceptor'. This dual function of sensing and transport is shared with other nutrient transporters in *S.cerevisiae* [12]. The subsequent activation of the Pi transporter system and maintenance of a stable intracellular Pi level is mediated by the phosphate-responsive signaling (PHO) pathway. This pathway is mainly regulated by Pho4, the transcriptional factor (TF); Pho80-Pho85, the cyclin dependent kinase (CDK) complex and Pho81, a cyclin dependent kinase inhibitor (CKI) (Fig. 2). Under Pi sufficient conditions, the nuclear localized Pho4 is phosphorylated by the Pho80-Pho85 CDK complex at multiple serine residues, resulting in disassociation from its cooperative TF Pho2 and its export from the nucleus into the cytosol [13]. When Pi is depleted, the conformation of Pho81, which constitutively associates with Pho80-Pho85 complex, is changed through interaction with inositol heptakisphosphate (IP7) [14] and therefore prevents the phosphorylation of Pho4 by Pho80-Pho85. The accumulated nuclear Pho4 interacts with Pho2 and activates the downstream Pi starvation induced (PSI) genes. A yeast genome wide analysis has revealed 22 genes that are transcriptionally regulated by this pathway, including those encoding high-affinity Pi transporters; Spl2, a negative regulator of low-affinity Pi transporters; secreted phosphatases; the vacuolar poly Phosphate (poly P) synthesis complex and Pho81 itself [15].

The yeast Pi transporter system consists of two high-affinity Pi transporters (Pho84, Pho89) and two low-affinity Pi transporters (Pho87, Pho90) that are localized in the plasma membrane, as well as a vacuolar membrane localized Pho91 [16] (Fig. 2). While the expression of Pho84 and Pho89 is highly induced by exposure to low Pi conditions, the low-affinity transporters are constitutively transcribed and are endocytosed for vacuolar degradation upon Pi depletion [17]. Although this vacuolar targeting process is carried out through distinct pathways for Pho87 and Pho90, either dependent or independent on Spl2, respectively, the N-terminal conserved SPX (**S**yg1-**P**ho81-**X**pr1) domain is strictly required [17], suggesting a potential protein interaction site within the SPX domain.

Upon Pi addition to the medium, the Pi transceptors Pho84 and Pho87 sense high environmental Pi and transport it across the plasma membrane [18]. Pi-induced activation of the protein kinase A (PKA) pathway in turn leads to the phosphorylation and ubiquitination of Pho84 itself, and eventually results in its internalization and degradation in the vacuole [19] (Fig. 2). Interestingly, Pinson *et al.* [20] showed that independently of intracellular Pi level, the low-affinity Pi transporters could regulate the expression of PSI genes in a Pho81 dependent manner, possibly through signaling pathway involving protein interaction at their N-terminal hydrophilic SPX domain.

During Pi uptake, the intracellular Pi content is also balanced by the inorganic polyphosphate (poly P) synthesis and vacuolar accumulation, which is accomplished by four Vacuolar Transporter Chaperone (Vtc) family members [15] (Fig. 2). Although Vtc4 has been identified as the polyP synthesizing enzyme [21], the fully functional Vtc complex requires the integrity of a heterotrimer Vtc1/2/4 or Vtc1/3/4, indicating the existence of protein-protein interactions between subunits, possibly through the SPX domain of Vtc2, 3 and 4. No Vtc member has been identified in plant or mammals, suggesting Pi storing in higher eukaryotic cells may require different participants. Pi starvation signals also trigger the scavenging of macromolecules for Pi remobilization. This procedure involves expression upregulation of secreted acid phosphatases (Pho5, Pho11, Pho12), vacuolar polyphosphatase (Phm5), glycerol phosphatase (Hor2) as well as the glycerophosphoinositol (GroPIns) / glycerophosphocholine (GroPCho) transporter (Git1) and glycerophosphodiester phosphodiesterase (Gde1), activity of which is also seen in bacterial, plant and mammalian cells [15,22-25].

Pi regulation in plants

Unlike bacteria and yeast, Pi homeostasis maintenance in plants requires different intracellular regulatory pathways in cells from varying tissue types, as well as intricate intercellular signaling and communication. As Pi sensing and signaling integration on the whole plant scale has already been extensively reviewed [26,27], this article will focus on the main intracellular signaling pathways (Fig. 3) and their comparison to the ones from single-celled organisms. In *Arabidopsis*,

the ER localized Low Phosphate Root 1 (LPR1, a multi copper oxidase) and Phosphate Deficiency Response 2 (PDR2, a P5 type ATPase) have been shown to play key roles in rhizosphere Pi sensing. LPR1 contributes to the reduced primary root cell length and PDR2 regulates stem cell differentiation and meristem activity through the expression modulation of SCARECROW (SCR) [28], whereas more general root architecture response is controlled by SIZ1 (SAP and Miz1, a SUMO-E3 ligase) dependent auxin regulation [29]. Notably, Pi sensing alone is sufficient to locally control the primary root growth as the non-metabolizable phosphite can also rescue the root meristem activity under low Pi conditions [30]. A minority of plants, mainly from the Proteaceae family, also produces clustered lateral roots (cluster roots, CRs) to increase the nutrient absorbing area as well as to secrete organic acids and acid phosphatase, in order to survive on the most Pi impoverished soils [31].

Low Pi sensing also triggers the regulation of multiple intracellular transcription factors [26] (Fig. 3), among which the MYB family member Phosphate Starvation Response 1 (PHR1) plays a critical role of regulating downstream PSI genes by binding to a P1BS (**P**HR**1**-**B**inding **S**ite) motif in their promoter regions [32]. The nucleus localized PHR1 is responsible for the Pi starvation induced upregulation of Phosphate Transporter 1 (PHT1), proposed Pi exporter PHO1;H1, regulatory microRNA399 (miRNA399) as well as the expression level changes of SPX-exclusive family member AtSPX1-4 [32-34]. Although PHR1 has previously been shown to be post-translationally controlled by SIZ1 through sumoylation, recent studies in both *Arabidopsis* and rice (*Oryza sativa*) have revealed that the activity of AtPHR1/OsPHR2 is also modulated by SPX1/2 through a competitive inhibition of PHR1 binding to P1BS in a Pi dependent manner [35-37]. Another study in rice also demonstrated under Pi sufficient condition, OsSPX4 can negatively regulate the OsPHR2 activity by retaining it in the cytoplasm as well as by interacting with the P1BS binding domain of nuclear localized OsPHR2 [38]. These findings suggest a possible intracellular Pi sensing function of SPX domain-containing proteins, although further evidence is needed to better understand the precise role of this SPX domain and the mechanism of Pi sensing and signaling.

Kinetic analysis in *Arabidopsis* has shown that multiple Pi transport systems are required especially in the presence of low environmental Pi [39]. After the first membrane Pi transporters were identified in *Arabidopsis* and later designated as the Phosphate Transporter 1 (PHT1) family members, other PHT1 members have been identified in *Arabidopsis* and other plants [40-42]. Members of the PHT1 family are generally Pi / H^+ co-transporters that belong to the major facilitator superfamily (MFS) and are each predicted to share a similar topological structure, comprising 2 bundles of 6 trans-membrane α-helices and both the C- and N-termini located on the cytosolic side of the membrane [43]. In *Arabidopsis*, only PHT1;1 and PHT1;4 are root specific Pi transporters and have been identified as homologues of yeast high-affinity Pi transporter Pho84, while the other seven PHT1 members are widely expressed in shoot and pollen [44,45], indicating a plant-wide Pi uptake and remobilization function of the PHT1 family. The expression level of *PHT1* genes is up- and down-regulated by PHR1, WRKY75 and MYB62,

respectively [46,47], whereas the homeostasis of PHT1 is post-translationally modulated by PHF1 (**PH**osphate transporter traffic **F**acilitator 1) through protein trafficking and membrane targeting [48,49] as well as by NLA and PHO2 through ubiquitination and subsequent endocytosis and vacuolar degradation [50] (Fig. 3).

It is of note that due to the sessile nature of plants, formation of a mutualistic arbuscular mycorrhizal (AM) symbiosis can greatly increase the plant Pi uptake from soil. Around 80% of plant species form this type of association. Although the entire mechanism involved in nutrients interchange between plants and mycorrhizal fungus remains to be elucidated, specific PHT1 family members have been demonstrated to play key roles in this essential plant Pi uptake process. Among these PHT1 transporters, two subfamilies have been classified, namely the subfamily III Pi transporters whose expression is enhanced by symbiosis and the subfamily I Pi transporters whose expression is specifically seen in symbiosis [51]. Although physically connected to the plasma membrane of cortical cells, the periarbuscular membrane harbors a distinct population of proteins that are critical for AM symbiosis. Recent study of *Medicago truncatula* MtPT4 has demonstrated establishment of this periarbuscular membrane specific protein pool is achieved by a transient expression and re-orientated secretion of Pi transporters in the colonized root cells [52]. Interestingly, study in monocotyledon plant rice (*Oryza sativa*) showed that although the Pi demands of mycorrhizal rice could be solely satisfied by OsPT11, another phylogenetically distant PHT1 member OsPT13 is also required for the integrated formation of AM symbiosis, indicating non-redundant functions of AM-specific Pi transporters [53].

The cell-to-cell-movement of Pi in multicellular organism requires both importers and exporters of this vital nutrient. An earlier isolated *Arabidopsis pho1* mutant exhibited deficient Pi accumulation in leaves but no detectable impairment in Pi root uptake rate or shoot xylem transfer, indicating a specific Pi xylem loading function of plasma membrane localized PHO1 [54]. The gene was identified via a map-based cloning strategy. Sequencing revealed that there are 10 *Arabidopsis* genes that encode proteins which share with PHO1 a conserved N-terminal SPX domain and a hydrophobic C-terminal EXS (**E**RRD 1-**X**PR1-**S**YG1) domain with six to eight predicted membrane-spanning segments [55]. Among the broadly expressed 11 *Arabidopsis* PHO1 family members, only PHO1 and PHO1;H1 are found to be responsible for Pi export, and their activity is under the regulation of E2-3 ligase PHO2 and the main transcription factor PHR1, respectively [56,57] (Fig. 3). AtPHO1;H4 is shown to control hypocotyl elongation under blue light [58] and the low Pi inducible AtPHO1;H10 is involved in the response to a number of biotic and abiotic stresses other than Pi starvation [59]. The study of PHO1 family members in both *Arabidopsis* and rice has further verified their Pi export function as well as their responsibility for Pi transfer from roots to shoots [60,61]. Recent investigation of XPR1, the closest mammalian homologue of PHO1, also demonstrated similar sub-cellular localization as well as Pi export mediating functions though the N-terminal SPX domain has been shown to be dispensable for Pi efflux [62,63]. However, in spite of the accumulating

evidence showing a Pi exporting function for PHO1 and its homologues, it is still unclear how this function is modulated intracellularly in response to changes in Pi levels. Although translocation of PHO1 from the endomembrane system to the plasma membrane has been observed upon high Pi infiltration [64], increased levels of XPR1 at the metazoan cell surface did not lead to elevated Pi export, indicating that additional factors may be required in mammalian cells [63]. While PHO1 mediates efflux from the cytosol to the apoplast, a recent study in *Arabidopsis* revealed that the tonoplast localized **V**acuolar **P**hosphate **T**ransporter 1 (VPT1) is responsible for the Pi efflux from cytosol into the vacuole for storage [65]. This finding has provided important information on plant vacuolar Pi sequestration and storage, whereas future work is needed to fully understand the mechanism of plant adaptation to variable Pi availability. One key step is to identify components that are involved in Pi release from the vacuole upon Pi starvation.

A mutant allele of the sucrose transporter gene *SUC2 (pho3)*, exhibits lower root acid phosphatase activity, low Pi content and other Pi-deficient symptoms along with a significant expression increase of the plastidic glucose 6-phosphate/phosphate translocator [66,67]. Mutations within the negative regulator of ethylene biosynthesis *Ethylene Overproduction 1* (*ETO1*) have also been found to lead to the altered expression of Pi starvation induced (PSI) genes and enhanced acid phosphatase (APase) production independently of the Pi supply [68]. These observations together demonstrate the involvement of other components such as sugar (reviewed by Hammond & White [69]) and phytohormones in the complicated signaling mechanisms of systemic Pi regulation in plants.

Conclusions

The Pi sensing and signaling systems in unicellular bacteria and yeast cells are better understood models of intracellular Pi regulation than their plant counterparts. However, similarities of Pi regulation patterns can be observed between the single cell and multi-cell systems. For instance, although not being orthologs, both the plant VPT1 and yeast VTC complex are responsible for transport of Pi into the vacuole for storage; whereas the plant high-affinity Pi transporters from the PHT1 family have been identified as homologues of *S. cerevisiae* Pi transceptor Pho84, and indeed many plant PHT1 members have been shown to be able to complement the *S. cerevisiae pho84* mutant [40,70]. Inositol phosphates (InsPs), which serve as inositol and phosphorus storage compounds in higher plants as well as signaling molecules, have also been demonstrated to play important roles in both yeast and plant Pi homeostasis maintenance [71]. In all three systems, the main intracellular Pi specific transcription factors (bacterial PhoB, yeast Pho4, plant PHR1) are regulated by their target gene products, suggesting similar feedback loops exist among different prokaryotic and eukaryotic organisms even though the components are not necessarily homologues or orthologues. Structural information on these main factors as well as

their interacting partners will help to gain a better understanding of Pi regulation pathways across all systems.

It is of note that in both yeast and plants many of the key players in Pi sensing and signaling all harbor a highly conserved N-terminal SPX domain (Table 1) and that this domain is absent from prokaryotes. Interestingly, previous studies have shown that this SPX domain is dispensable for any catalytic activity and transport function [21,63], but acts as an auto-inhibitor of its host proteins [72], suggesting this domain is functioning as a regulatory domain by physical interaction with other protein domains. However, in spite of the accumulating evidence of the SPX domain being involved in protein-protein interactions, it is still unknown whether any of the well-conserved residues are of significant importance to specific interactions or whether these interactions could be regulated by intracellular Pi signals through a Pi sensing function of this SPX domain. Therefore, structural information is urgently needed for the conserved SPX domain to provide insights into its *in vivo* function in Pi homeostasis regulation. Identification of more Pi sensing elements as well as SPX domain-containing proteins interacting partners will also help to obtain a fuller view of the complicated eukaryotic Pi regulation pathway network.

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Table1. Pi regulation related SPX domain-containing proteins in Yeast (*Saccharomyces cerevisiae*), *Arabidopsis* and Rice (*Oryza sativa*).

Species	Component	Function
Saccharomyces cerevisiae (Yeast)	Pho81	Cyclin-dependent kinase inhibitor: Interacts with IP7, inhibits the activity of the Pho80–Pho85 complex
	Pho87	Low affinity Na^+ /Pi transporter: Pi sensing and transporting under high Pi condition
	Pho90	Low affinity Na^+ /Pi transporter: Pi transporting under high Pi condition
	Pho91	Vacuolar Pi transporter: Pi transporting from the vacuole to the cytosol
	Vtc2/3/4	Vacuolar Transporter Chaperon Complex components: Vacuolar polyphosphate accumulation, membrane trafficking, microautophagy and non-autophagic vacuolar fusion
	Gde1	Glycerophosphocholine phosphodiesterase: Hydrolyzes GroPCho to choline and glycerolphosphate for Pi scavenging from macromolecules
Arabidopsis	SPX1/2	SPX-exclusive family members: Pi-dependent inhibitors of the activity of PHR1 & interact with PHR1
	SPX3	SPX-exclusive family member: Regulates plant adaptation to Pi starvation & negatively regulates some PSI genes
	PHO1/PHO1;H1	SPX-EXS family members: Pi exporting and xylem loading
	SPX-MFS3	SPX-MFS family member: Vacuolar Phosphate Transporter 1, Pi transport into the vacuole
	NLA	SPX-RING family member: Pi regulation under N limited condition & anthocyanin synthesis regulation
Oryza sativa (Rice)	SPX1/2	SPX-exclusive family members: Pi-dependent inhibitors of the activity of PHR2 & interacts with PHR2
	SPX3/5	SPX-exclusive family member: Negatively regulate root-to-shoot Pi translocation & functional repressors of PHR2
	SPX4	SPX-exclusive family member: Negative regulator of PHR2 & reduces the nucleus targeting of PHR2
	PHO1;2	SPX-EXS family member: Pi transport from root to shoot
	SPX-MFS1	SPX-MFS family member: Pi transport and remobilization in leaves

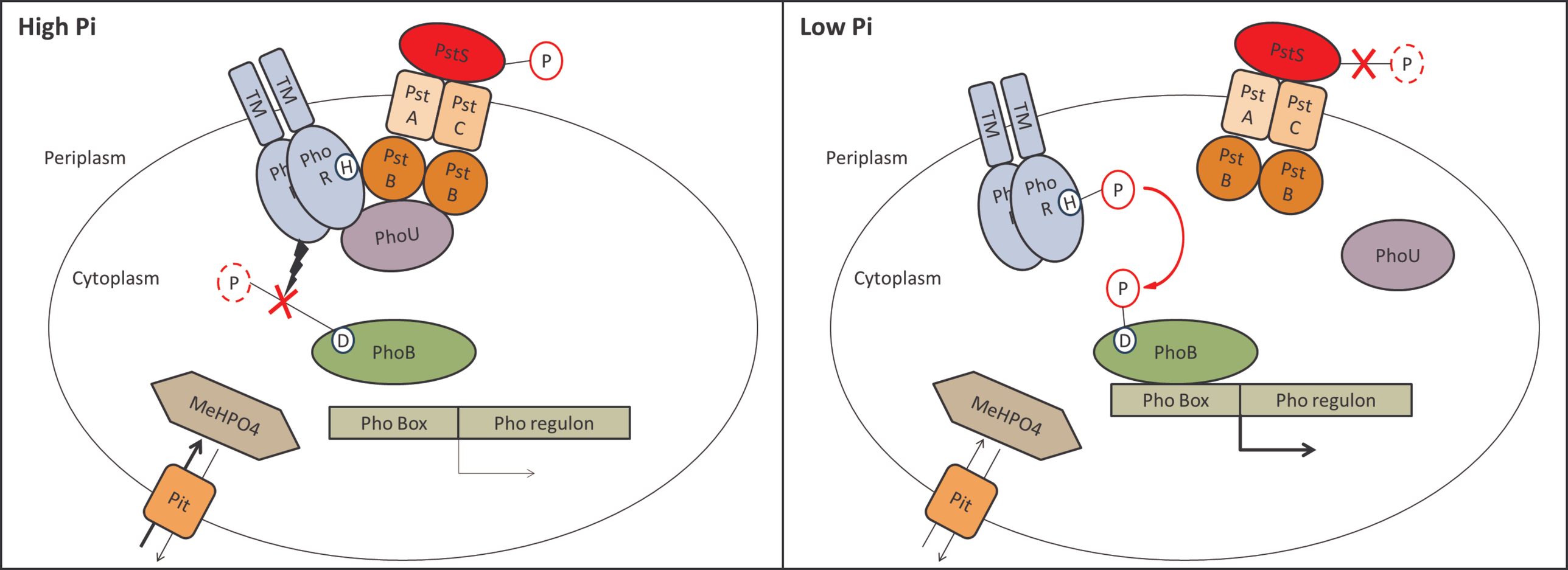
Figure legends:

Fig1. Schematic diagram of Pi regulation pathways in *E. coli*. Under high Pi conditions, binding of Pi to PstS results in inhibition of histidine kinase PhoR via the formation of a PhoR-PhoU-PstB protein complex, resulting in dephosphorylation and deactivation of the transcription factor PhoB. The low-affinity Phosphate inorganic transporter (Pit) system also mediates metal-phosphate complexes (MeHPO4) transport across the plasma membrane. Under low Pi conditions, PhoB is activated by phosphorylation on an aspartate residue (D) by the auto-

phosphorylated histidine kinase PhoR and binds to specific DNA targets through the Pho box thereby allowing the activation of downstream Pho regulon genes.

Fig2. Schematic diagram of Pi regulation pathways in *S. cerevisiae*. Under high Pi conditions, Pho4 is phosphorylated on multiple serine residues (S) by the Pho80-Pho85 CDK complex and exported from the nucleus into the cytosol. The low-affinity Pi transporters Pho87 and Pho89 are responsible for Pi uptake and high-affinity Pi transporter Pho 84 is internalized and degraded. Under low Pi conditions, interaction with IP7 changes the conformation of Pho81 and prevents the phosphorylation of Pho4 by Pho80-Pho85. Accumulated nuclear Pho4 in concert with Pho2 activates the downstream Pi starvation induced (PSI) genes and leads to the upregulation of high-affinity Pi transporters as well as the internalization and degradation of low-affinity Pi transporters. Low Pi also triggers the formation of Vtc complex to increase the vacuolar accumulation of inorganic polyphosphate (polyP), as well as polyP hydrolysis by vacuolar polyphosphatase Phm5 and Pi transport from vacuole to cytosol by Pho91. Pi scavenging from macromolecules is also activated during Pi starvation, resulting in the import of glycerophosphoinositol (GroPIns) and glycerophosphocholine (GroPCho) through Git1 and the hydrolysis of GroPCho by Gde1.

Fig3. Schematic diagram of Pi regulation pathways in *Arabidopsis* (Arrowheads and blunt-ends show upregulation and downregulation, respectively). Limited environmental Pi level is sensed through the root tip by LPRs and PDR2, introducing root architecture reprogramming. The low Pi signal activates the main intracellular transcription factor PHR1 and its downstream PSI genes to increase the Pi uptake from soil (PHT1 family) as well as Pi relocation in the plant (PHO1 family and some PHT1 family members). Activities of transcription factor ZAT6, WRKY75, MYB62 and bHLH32 are also upregulated and downregulated, respectively, in response to Pi starvation. Non-coding RNAs (miR399, miR827) mediate post-transcriptional regulation of E2-3 ligase PHO2 and E3 ligase NLA which in turn regulate PHT1 protein levels as well as their intracellular protein trafficking via PHF1.


High Pi
PstS
P
Pst A
Pst C
TM
TM
Pho R
H
Pst B
Pst B
PhoU
Periplasm
Cytoplasm
P
D
PhoB
MeHPO4
Pit
Pho Box
Pho regulon
Low Pi
PstS
P
Pst A
Pst C
TM
TM
Pho R
H
Pst B
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P
PhoU
Periplasm
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P
D
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Pit
Pho Box
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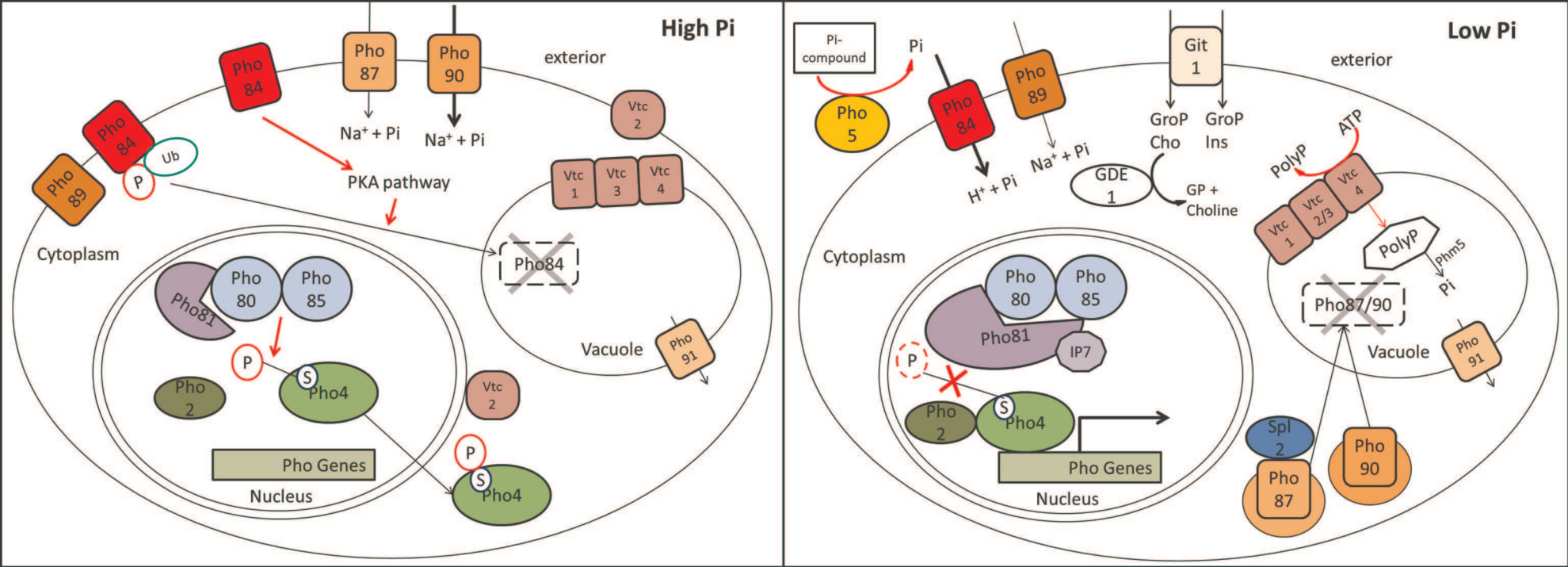
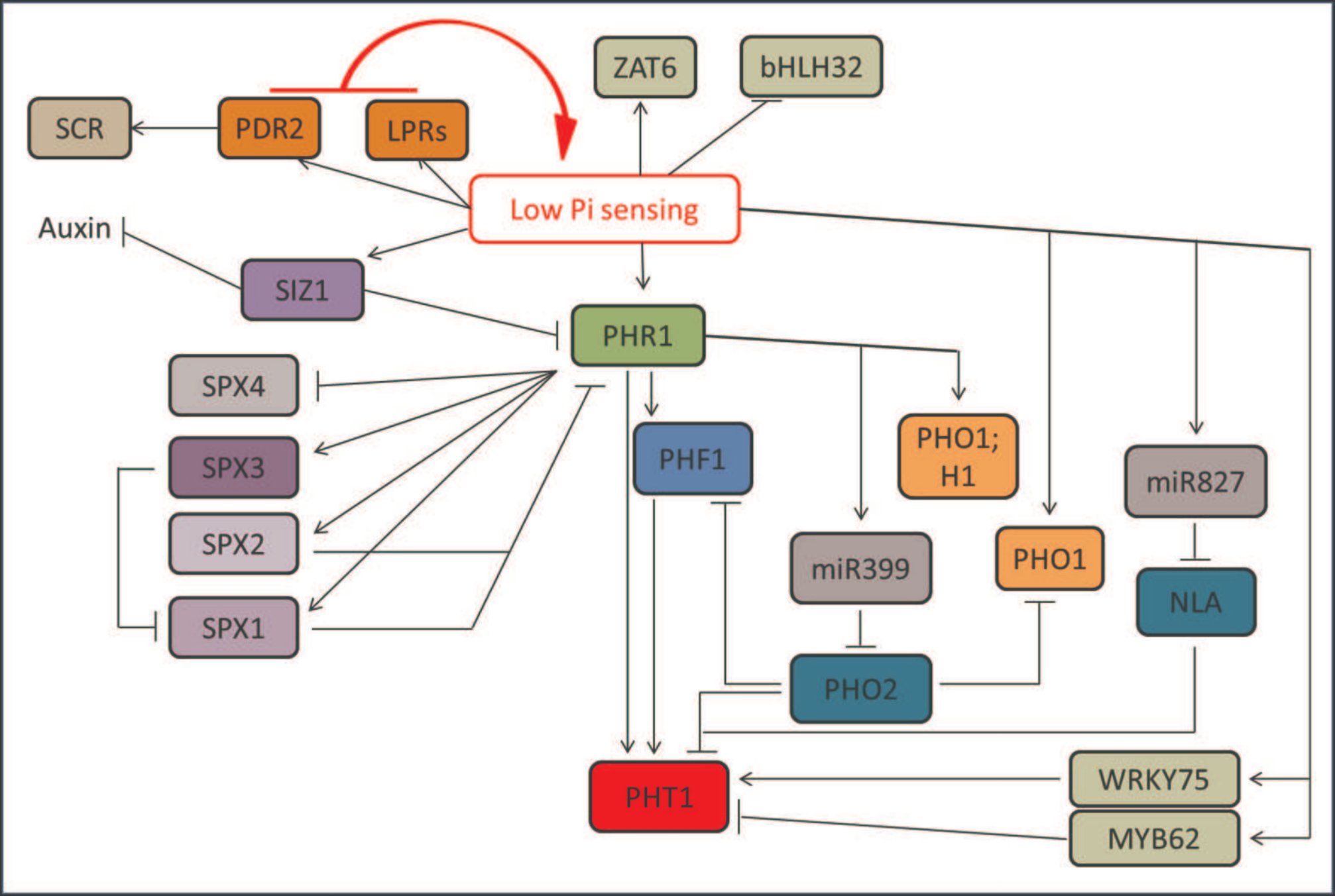

High Pi
exterior
Pho 84
Pho 87
Pho 90
Na+ + Pi
Na+ + Pi
Vtc 2
Vtc 1
Vtc 3
Vtc 4
Pho 89
Pho 84
Ub
P
PKA pathway
Cytoplasm
Pho84
Pho81
Pho 80
Pho 85
P
S
Pho4
Pho 2
Pho Genes
Nucleus
Vacuole
Pho 91
Vtc 2
P
S
Pho4
Low Pi
Pi-compound
Pho 5
Pi
Pho 84
Pho 89
H+ + Pi
Na+ + Pi
Git 1
GroP Cho
GroP Ins
GDE 1
GP + Choline
exterior
ATP
PolyP
Vtc 4
Vtc 2/3
Vtc 1
PolyP
Phm5
Pi
Cytoplasm
Pho 80
Pho 85
Pho81
IP7
P
S
Pho4
Pho 2
Pho Genes
Nucleus
Pho87/90
Vacuole
Pho 91
Spl 2
Pho 87
Pho 90


ZAT6
bHLH32
SCR
PDR2
LPRs
Low Pi sensing
Auxin
SIZ1
PHR1
SPX4
SPX3
SPX2
SPX1
PHF1
PHO1; H1
miR827
miR399
PHO1
NLA
PHO2
PHT1
WRKY75
MYB62