

Pumilio RNA Binding Protein in Plants

Subjects: Agriculture, Dairy & Animal Science

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RNA binding proteins (RBPs) have been known to control target genes. One type of protein, Pumilio (Pum)/Puf family RNA binding proteins, show a specific binding of 3' untranslated region (3' UTR) of target mRNA and function as a post-transcriptional/translational regulator in eukaryotic cells. Plant Pum protein is involved in development and biotic/abiotic stresses.

Keywords: RNA binding protein ; Pumilio ; translational modification ; RNA biomolecules

1. Protein–Protein Interaction in Plant Pum

Pum-HD has a dual function. The inner surface of Pum-HD binds to the conserved 'UGUX₃₋₅UA' motif in the 3' UTR regions of the target gene. Interestingly, the outer surface of Pum-HD also permits protein–protein interactions with diverse proteins ^{[1][2]}. The yeast Pum protein, PUF5p, binds to and represses *HO* mRNA, which encodes a DNA endonuclease via binding to the Pumilio-RNA binding motif. Pop2p, which encodes a component of the Ccr4p-Pop2p-Not deadenylase complex, is required for PUF5p-mediated *HO* mRNA decay ^[3]. It affects both mRNA stability and translational efficiency ^{[4][5]}. Thus, Pums do not work alone to repress the target mRNA. Furthermore, yeast PUF5p and Pop2p interaction is evolutionally conserved in *C. elegans* PUF8 and *H. sapiens* (PUM1) ^[3]. Arabidopsis Pum, APUM5, also interacts with plant Ccr4 homologues ^[6]. Sometimes, yeast PUF5p does not require Ccr4-Pop2 deadenylase when PUF5p responds to DNA replication stress ^[7]. Thus, plant Pum function could be associated with deadenylase-dependent and -independent pathways via formation, as with Ccr4-Pop2-NOT mRNA deadenylase complexes.

Human PUM2 can make a complex with Argonaute (Ago) miRNA-binding protein, and then, the PUM2-Ago heterodimer is associated with the core translational elongation factor (eEF1A). This complex inhibits eEF1A GTPase activity and translational elongation ^[8]. These results imply that plant Pum could be associated with the Ago-eEF1A protein complex.

2. The Role of Pum in Plant Development

Pum proteins play important roles during development, differentiation and cell cycle regulation in various organisms ^{[9][10]}. APUM1 to APUM6 are specifically associated with shoot stem cell maintenance genes such as *WUSCHEL* (*WUS*) and *CLAVATA1* (*CLV1*) ^{[11][12][13]}. Like the mammalian system, human PUM2 is expressed in embryonic stem cells and could affect germ cell development ^[14]. Mammalian PUM1 and PUM2 regulate cell cycle inhibitor Cdkn1b via translational control ^[15]. *C. elegans* Pum, PUF8, might function as a repressor of the stem cell proliferative fate via control of GLP-1/Notch signaling in germline cells ^[16]. Likewise, plant Pum could affect a variety of development stages. Gene expressions of *APUM23* and *APUM24* were shown to be continuously expressed in all the developmental stages and were enhanced at the seed stage (**Figure 2A**). *APUM23* affects pre-ribosomal RNA processing in the nucleolus and the *apum23* mutant showed a developmental defeat phenotype ^[17]. It is possible that *APUM24* could function in the ribosomal RNA processing because *APUM24* predominantly localizes to the nucleolus ^[18]. *APUM5* and *APUM6* were highly abundant at developmental stages (**Figure 3**). However, these mutants did not show abnormal developmental phenotypes. *APUM1* to *APUM6* have a similar protein structure and might have the same redundancy. In the future, this will be studied for genetic crossover with *APUM1* to *APUM6* mutants. In rice, at least four rice *Pum* genes are highly expressed at the vegetative stage (**Figure 2B**). Plant Pum proteins could play a role in development and differentiation.

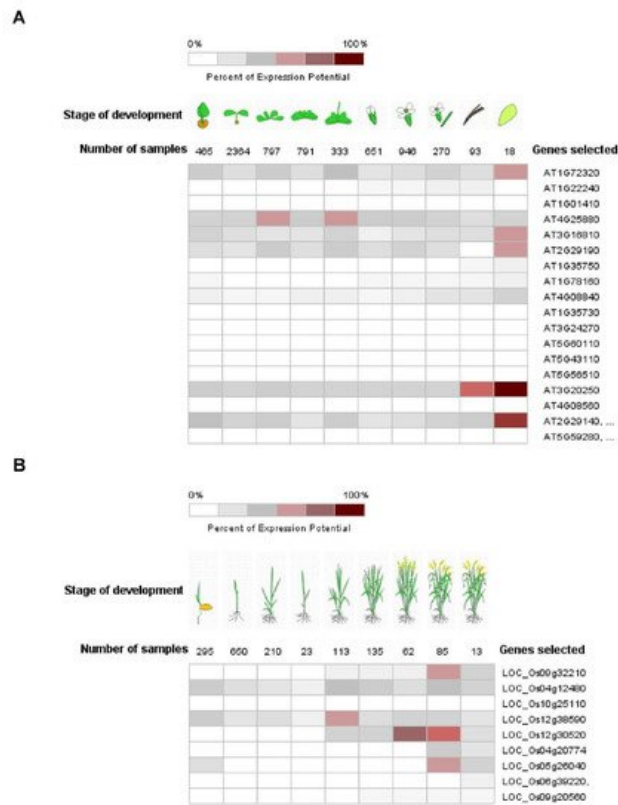


Figure 2. Expression pattern of Arabidopsis and rice *Pum* in development. **(A,B)** Expression pattern of Arabidopsis and rice *Pum* was analyzed at developmental stage using the Genevestigator (<https://www.genevestigator.com> accessed on 13 November 2021). Expression levels of Arabidopsis and rice *Pum* vary under developmental stages. Transcript levels were assessed using microarray data available from Genevestigator.

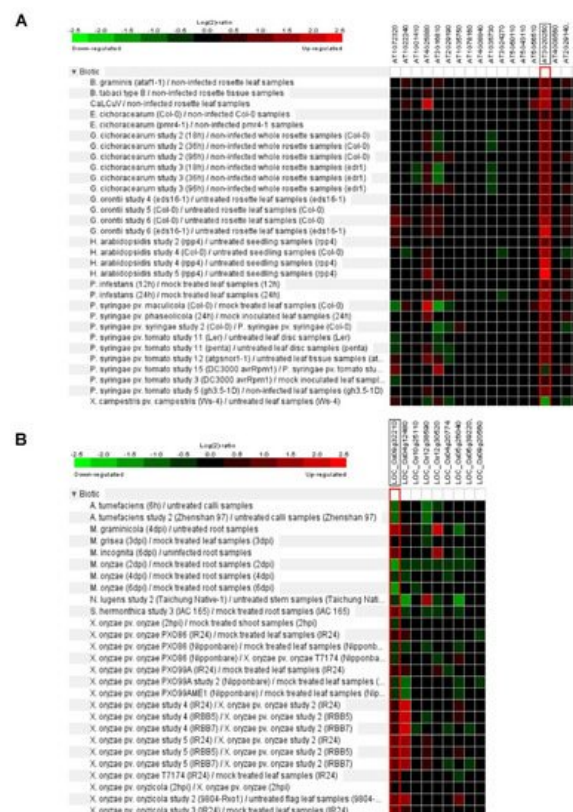


Figure 3. Biotic stress response of Arabidopsis and rice *Pum*.

3. Plant *Pum* Function in Biotic Stress

In Arabidopsis, some putative target RNAs of *Pum* were identified by yeast in a three-hybrid system. These genes contained Pumilio RNA binding motifs at the 3' UTR regions [11]. For example, *Responsive to dehydration 19* (*RD19*) and *Plant homeodomain* (*PHD*) containing protein (At3g63500) were isolated [11]. *RD19* encodes cysteine protease and is involved in resistance and gene-mediated immunity via interaction with *Ralstonia solanacearum* type III effector PopP2

4. Plant Pum Function in Abiotic Stress

A

Color scale: -2.0 (green) to 2.0 (red)

Legend: *longiracata* (green), *longiracata* (red)

Gene list (top):

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Figure 4. Differential expression patterns of *Pum* genes in Arabidopsis and rice in response to abiotic stresses or phytohormones. **(A)** Expression pattern of Arabidopsis *Pum* was analyzed in abiotic stresses (cold, drought, heat, hypoxia, osmotic and salt) and phytohormones (ABA and brassinosteroid) treatments using Genevestigator. **(B)** Expression pattern of rice *Pum* was analyzed in abiotic stresses (anaerobic, cold, drought, heat, and salt) and phytohormones (gibberellin, auxin, and cytokinin) treatments using Genevestigator. Arabidopsis and rice *Pum* transcript levels were assessed using microarray data available from Genevestigator. Expressions patterns were searched in a range of abiotic stress conditions in this database. Red colors indicate genes that were upregulated relative to the control for a given treatment, and green colors indicate genes that were downregulated relative to the control.

5. The Multifunctional Plant Pum Protein and Crop Engineering

The translational control of gene expression is an important step for the survival of plants. Pum proteins evolutionally conserve the canonical Pum-HD RNA binding domain in most of the eukaryotes. This Pum-HD confers target RNA specificity via direct binding and functions as a protein–protein interaction platform. Sometimes, Pum acts as a translational activator. Until now, plant Pum functions were not well known. The plant *Pum* family contains the highest number of members, about twenty, compared with the mammalian *Pum* family. This implies that many plant Pum proteins might have plant-specific functions and could have plant-specific targets. From the in silico data analysis, it is shown that plant Pums could be associated with a variety of signaling pathways. This regulation could be more complicated because plant hormones exhibit crosstalk between different plant hormones. A better understanding of the plant Pum regulation mechanism would help innovate new strategies to improve crop plant engineering. Thus, it would be interesting to identify the putative RNA targets and generate customized Pum-HD proteins for agriculture.

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