Division of Biochemistry
– Molecular Biology –

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Prof
AOYAMA, Takashi
(D Sc)

Assoc Prof
TSUGE, Tomohiko
(D Sc)

Assist Prof
KATO, Mariko
(D Agr)

Students

AKAGI, Chika (D3)
HAGHIR, Shahrzad (D2)

Scope of Research

This laboratory aims at clarifying molecular bases of regulatory mechanisms for plant development, especially plant morphogenesis, with techniques of forward and reverse genetics, molecular biology, and biochemistry. Current major subjects are: 1) phospholipid signaling in cell morphogenesis, 2) the transcriptional network for cytokinin responses, 3) COP9 signalosome modulating signal transduction in the nuclei, and 4) the endoreduplication cell cycle in cell differentiation.

KEYWORDS

Morphogenesis
Phospholipid Signaling
mRNA Processing
Signal Transduction
COP9 Signalosome

Recent Selected Publications

Watari, M.; Kato, M.; Blanc-Mathieu, R.; Tsuge, T.; Ogata, H.; Aoyama, T., Functional Differentiation among the Arabidopsis Phosphatidylinositol 4-Phosphate 5-Kinase Genes *PIP5K1*, *PIP5K2* and *PIP5K3*, *Plant Cell Physiol.*, **63,** 635-648 (2022).

Zhang, X.; Nomoto, M.; Garcia-León, M.; Takahashi, N.; Kato, M.; Yura, K.; Umeda, M.; Rubio, V.; Tada, Y.; Furumoto, T.; Aoyama, T.; Tsuge, T., CFI 25 Subunit of Cleavage Factor I Is Important for Maintaining the Diversity of 3′ UTR Lengths in *Arabidopsis thaliana* (L.) Heynh., *Plant Cell Physiol.*, **63,** 369-383 (2022).

Shimamura, R.; Ohashi, Y.; Taniguchi, Y. Y.; Kato, M.; Tsuge, T.; Aoyama, T., Arabidopsis PLDζ1 and PLDζ2 Localize to Post-Golgi Membrane Compartments in a Partially Overlapping Manner, *Plant Mol. Biol.*, **108,** 31-49 (2021).

Kuroda, R.; Kato, M.; Tsuge, T.; Aoyama, T., Arabidopsis Phosphatidylinositol 4-phosphate 5-kinase Genes *PIP5K7*, *PIP5K8*, and *PIP5K9* Are Redundantly Involved in Root Growth Adaptation to Osmotic Stress, *Plant J.*, **106,** 913-927 (2021).

Aki, S. S.; Yura, K.; Aoyama, T.; Tsuge, T., SAP130 and CSN1 Interact and Regulate Male Gametogenesis in *Arabidopsis thaliana*, *J. Plant Res.*, **134,** 279-289 (2021).

Genetic Research of Plant PIP5K Genes

Phosphatidylinositol (4,5)-bisphosphate [PtdIns(4,5)P_2], a phosphoinositide serving as a lipid signal interacting with its effector proteins, modulates various cellular processes including cytoskeletal organization, membrane trafficking, and signal transduction for gene expression. Although the metabolic pathways of phosphoinositides are elaborately linked to one another, phosphatidylinositol 4-phosphate 5-kinase (PIP5K), which produces PtdIns(4,5)P_2 by phosphorylating PtdIns(4)*P*, is thought to be a key enzyme responsible for the spatiotemporal pattern of PtdIns(4,5)P_2 in higher plant cells. Higher plants encode a large number of PIP5Ks compared with animals and fungi. The model plant *Arabidopsis thaliana* encodes eleven PIP5Ks. Of these, nine can be classified into three subgroups – PIP5K1–3, PIP5K4–6, and PIP5K7–9 – belonging to different type B clades while PIP5K10 and PIP5K11 are classified as type A PIP5Ks. Genetic and molecular biological studies have revealed that the PIP5K4–6, PIP5K7–9 and PIP5K10–11 subgroup genes redundantly function in pollen tube growth, cell adaptation to stressful conditions, respectively, while *PIP5K10* and *PIP5K11* are exclusively expressed in pollen and thought to be redundantly involved in actin cytoskeletal reorganization during pollen tube growth.

With regard to the PIP5K1–3 subgroup genes, a common function to all genes has not been found so far. The loss of *PIP5K2* function affects auxin-related phenomena including root gravitropism and lateral root development. The *pip5k1pip5k2* double mutation causes sever dwarfism accompanied by sieve-element defects. In contrast, *PIP5K3*, which produces a protein localized to the plasma membrane at elongating root hair apices and sites where root hair bulges are about to form, is involved in root hair elongation. Although PIP5K3 belongs to a clade closely related with but distinct from that of PIP5K1 and PIP5K2 in the molecular phylogenetic tree of plant PIP5Ks, it is unknown how the PIP5K1–3 subgroup genes differ in their expression patterns and protein functions.

We performed comparative analyses of the PIP5K1–3 subgroup genes to determine their conserved and/or differentiated functions. Genetic analysis revealed that *PIP5K1* and *PIP5K3* have distinct functions – total plant growth and root hair elongation, respectively – whereas *PIP5K2* redundantly has both functions. This pattern of functional redundancy well coincides with the overlapping pattern of their promoter activities. In transformation rescue experiments with promoter-swapped transgenes, PIP5K1 could completely substitute for PIP5K3 in terms of protein functions (Figure 1), but only partial substitution could be achieved in the reverse case (Figure 2). Phylogenetic analysis of angiosperm type B PIP5Ks revealed that PIP5K3 orthologs have evolved faster than have PIP5K1/2 orthologs. These findings suggest that during the evolution of the PIP5K1–3 subgroup genes, *PIP5K3* differentiated to specialize in promoting root hair elongation and lost some of the ancestral protein-encoded functions conserved in PIP5K1 and PIP5K2, whereas *PIP5K1* and *PIP5K2* differentiated from each other only in their promoter-directed expression patterns.

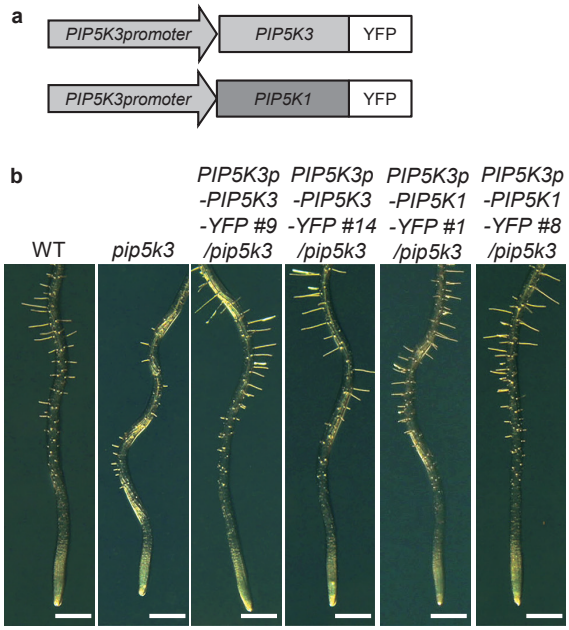

Figure 1. Rescue of the short-root-hair phenotype in *pip5k3* by *PIP5K3p-PIP5K1-YFP*. (a) Structures of the transgene *PIP5K3p-PIP5K3-YFP* and the promoter-swapped transgene *PIP5K3p-PIP5K1-YFP* are schematically illustrated. (b) Primary roots at 5 days after germination are shown for the wild type (WT), *pip5k3*, and the transformation lines harboring *PIP5K3p-PIP5K3-YFP* (lines #9 and #14) or *PIP5K3p-PIP5K1-YFP* (lines #1 and #8) in the *pip5k3* background. Bars = 1 mm

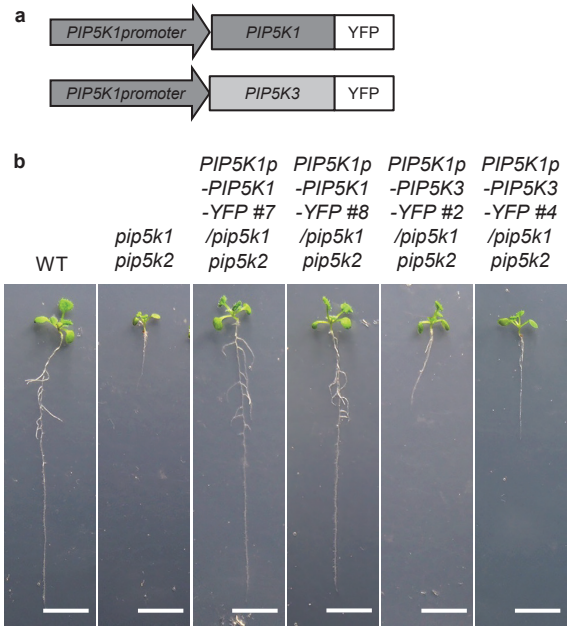

Figure 2. Partial rescue of the severe dwarf phenotype in *pip5k1pip5k2* by the promoter-swapped transgene *PIP5K1p-PIP5K3-YFP*. (a) Structures of the transgene *PIP5K1p-PIP5K1-YFP* and the promoter-swapped transgene *PIP5K1p-PIP5K3-YFP* are schematically illustrated. (b) Seedlings at 10 days after germination are shown for the wild type (WT), *pip5k1pip5k2*, and the transformation lines harboring *PIP5K1p-PIP5K1-YFP* (lines #8 and #7) or *PIP5K1p-PIP5K3-YFP* (lines #2 and #4) in the *pip5k1pip5k2* background. Bars = 1 mm