

論文内容の要旨

Studies of vacuolar trafficking pathways regulated by RAB5 and RAB7
in *Arabidopsis thaliana*

(シロイヌナズナにおける RAB5 と RAB7 が制御する
液胞輸送経路の研究)

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RAB GTPases regulate the tethering and fusion of transport vesicles to target membranes in membrane trafficking by acting as a molecular switch, cycling between GDP- and GTP-bound states. RAB5 is a member of the RAB GTPase family, the members of which have been shown to perform various functions in the endocytic pathway, including the regulation of endosomal fusion and motility in animal cells. RAB5-mediated endosomal trafficking has also been found to play important roles in various higher-order plant functions, which include the regulation of the polar transport of auxin and responses to environmental conditions. The regulatory mechanisms and functions of plant RAB5 have also been investigated at the molecular and cellular levels.

However, the significance of RAB5 activity at the tissue and organ levels has hardly been investigated thus far. In the present study, I examined the effect of a mutation in *VPS9a*, which encodes the sole guanine nucleotide exchange factor for all RAB5s in the vegetative stages of *Arabidopsis thaliana*. I found that multiple developmental processes were impaired in the mutant plants, including the growth and pattern formation of the roots and establishment of auxin maxima. My results indicate that RAB5 plays distinctive pivotal roles in the development of plants.

The multifunctional vacuole is the largest organelle in plant cells, and many proteins are transported to and stored in this organelle; thus, the vacuole has great physiological and agronomical importance. However, the molecular mechanism and regulation of plant vacuolar traffic remain largely unknown. In this study, I demonstrate that multiple vacuolar trafficking pathways operate in plants. RAB5 and RAB7 are evolutionarily conserved subfamilies of Rab GTPase, whose animal and yeast counterparts regulate vacuolar/endosomal trafficking in a sequential manner. Functional analyses of a putative activating complex for RAB7 indicated that this complex is responsible for maturation from RAB5- to RAB7-positive endosomes. Moreover, these machinery components are recruited to a more complex trafficking network. Mutations in RAB5 and RAB7 conferred counteracting effects on the *vti11*

mutant, which is defective in a vacuolar trafficking pathway. Furthermore, impairment of RAB5- and RAB7-dependent pathways differentially affected the transport of distinctive cargos. These results indicate that plants have developed a complex vacuolar transport system distinct from that of non-plant systems by assigning evolutionarily conserved machinery to unique trafficking pathways. These pathways provide a fundamental basis for plant development at the cellular and higher-ordered levels.