Since plants spread their roots in the ground, they must survive in a given environment. To adapt to the environment, they positively utilize environmental changes in the life cycle as important signals that are necessary for their survival. Plant cells can induce, degenerate and differentiate their organelles to adapt to environmental changes. The flexibility of plant organelles is the basis of the strategy for environmental adaptation in plants.

The aim of this division is to clarify the molecular mechanisms underlying the induction, differentiation, and interaction of organelles, and to understand the integrated function of individual plants through organelle dynamics.

The Scientific Research on Innovative Areas “Environmental sensing of plants: Signal perception, processing and cellular responses” was started to clarify the molecular mechanisms underlying organelle differentiation and interaction.

I. Reversible transformation of plant peroxisomes

Dramatic metabolic changes that underlie the shift from heterotrophic to autotrophic growth occur in the greening of seed germination. Accompanying these metabolic changes are the functional transformations of many constitutive organelles. Etioplasts differentiate into chloroplasts while mitochondria acquire the ability to oxidize glycine. Glyoxysomes, which are peroxisomes engaged in the degradation of reserve oil stored in oil body (Figure 1) via β-oxidation and the glyoxylate cycle, are transformed into leaf peroxisomes that function in several crucial steps of photorespiration. After the functional transition of glyoxysomes to leaf peroxisomes during the greening of pumpkin cotyledons, the reverse transition of leaf peroxisomes to glyoxysomes occurs during senescence. Gene expression, alternative splicing, protein translocation and protein degradation control the functional transformation between glyoxysomes and leaf peroxisomes.

II. Transcriptomics, proteomics and phenomics of plant peroxisomes

Enzymes localized in plant peroxisomes are synthesized in the cytosol and function after their post-translational transport into peroxisomes. Almost all of the peroxisomal matrix proteins contain one of two targeting signals (PTS1 and PTS2) within the molecules. PTS1 is a unique tripeptide sequence found in the carboxyl terminus of the mature proteins. In contrast, PTS2 is involved in a cleavable amino terminal presequence of peroxisomal proteins that are synthesized as a precursor protein with larger molecular mass.

We identified 256 gene candidates of PTS1- and PTS2-containing proteins and another 30 genes of non-PTS-containing proteins from the Arabidopsis genome. Custom-made DNA microarrays covering all these genes were used to investigate expression profiles of the peroxisomal genes in various organs. They revealed that peroxisomes in root cells play a pivotal role in polyamine catabolism. We also made a two-dimensional protein map of glyoxysomes and leaf peroxisomes isolated from Arabidopsis and soybean. Peptide MS fingerprinting analyses allowed us to identify novel peroxisomal membrane proteins, i.e. voltage-dependent anion-selective channel and adenine nucleotide carrier 1 (PNC1). We also found that peroxisomal membrane ATP-binding cassette transporter promotes seed germination by inducing pectin degradation under the control of abscisic acid signaling. The overall results provide us with new insights into plant peroxisomal functions.

Bioinformatic analysis of the Arabidopsis genome predicted the presence of 15 kinds of genes, called PEX genes, for peroxisomal biogenesis factors. We demonstrated that PEX5 and PEX7 form a cytosolic receptor complex and recognize PTS1- and PTS2-containing proteins, respectively. PEX14 is a peroxisomal membrane docking protein that captures the

![Figure 1. Oil body visualized by oleosin-GFP. (A) Five-day-old dark grown seedlings expressing oleosin-GFP. (B) Numerous oil bodies in the cell of dry seed. (C) High-resolution image of aggregated oil bodies. Bar = 5 μm.](image-url)
receptor-cargo complex. We also comprehensively investigated whether or not these predicted PEX genes function in peroxisome biogenesis by generating knock-down mutants that suppress PEX gene expression by RNA-interference. Phenotypes of these mutants allowed us to identify the functional PEX genes, which can be classified into two groups: PEX genes regulating for peroxisomal protein import and PEX genes regulating for peroxisomal morphology. We continue to investigate the detailed molecular functions of other PEX genes. Of these, we recently proposed that function of PEX7 is maintained by a quality control mechanism involving RabE1c.

III. Identification of novel components essential for peroxisome biogenesis

To better understand peroxisome biogenesis and functions, we isolated a number of Arabidopsis mutants having aberrant peroxisome morphology (apem mutants) based on them having a different pattern of GFP fluorescence from the parent plant, GFP-PTS1, in which peroxisomes with normal sizes and numbers can be visualized with GFP.

Of these apem mutants, APEM1 gene (whose defect causes the elongation of peroxisomes and mitochondria) encodes dynamin-related protein 3A, which is involved in division of both organelles. APEM2 and APEM4 (whose defects cause a decrease in the efficiency of protein transport) were revealed to encode proteins homologous to PEROXIN 13 (PEX13) and PEX12, respectively, and both proteins are responsible for protein translocation on peroxisomal membranes. APEM9 is the plant-specific PEX that has a role in tethering the PEX1-PEX6 complex on peroxisomal membranes. In addition, we found that APEM3 encodes Peroxisomal membrane protein 38, and that its defect causes enlargement of peroxisomes (Figure 2).

We are currently characterizing other apem mutants. From these analyses, we will be able to identify the components responsible for peroxisome biogenesis and functions, and to address the mechanism at the molecular level.

IV. ER derived organelles for protein storing and defense strategy

Plant cells develop various types of endoplasmic reticulum (ER)-derived structures with specific functions. ER bodies are ER-derived compartments observed in Arabidopsis. They are rod-shaped structures surrounded by ribosomes, and are widely distributed in the epidermal cells of whole seedlings. Undamaged rosette leaves have no ER bodies, but accumulate ER bodies after wounding or jasmonic acid treatment. This suggests that ER bodies function in the defense against herbivores. ER bodies in seedlings include β-glucosidase PYK10. When plant cells are damaged, PYK10 forms large protein aggregates. The aggregate formation increases glucosidase activity, possibly producing toxic products. Arabidopsis nai1 mutants have no ER bodies in the entire plant and do not accumulate PYK10. NAI1 encodes a transcription factor and regulates the expression of PYK10 and NAI2. The Arabidopsis nai2 mutant has no ER bodies and reduced accumulation of PYK10. NAI2 encodes a unique protein that localizes to the ER body. We found that the membrane protein of ER body 1 (MEB1) and MEB2 are integral membrane proteins of the ER body. MEB1 and MEB2 have homology to yeast iron/manganese transporter CCC1. Heterologous expression of MEB1 and MEB2 in yeast ccc1 mutant suppresses the iron toxicity (Figure 3), indicating that MEB1 and MEB2 are iron/manganese transporters. These results suggest that the ER body has specific membrane proteins that are involved in defense against metal stress as well as pathogens and herbivores. NAI2 deficiency relocates MEB1 and MEB2 to the ER network. These findings indicate that NAI2 is a key factor that enables ER body formation. We are now investigating the function of NAI2 on ER body formation by heterologously expressing it in onion and tobacco cells.

V. Roles of molecular chaperones on cell differentiation

Molecular chaperones are cellular proteins that function in the folding and assembly of certain other polypeptides into oligomeric structures. We have found that HSP90 inhibitor induces genes with heat shock response element (HSE)
motifs in their promoters, suggesting that heat shock transcription factor (HSF) is involved in the response. Arabidopsis HSFs interacted with HSP90.2. Thus, it appears that in the absence of heat shock, HSP90 actively suppresses HSF function. During heat shock, HSP90 is transiently inactivated, which leads to HSF activation. These data indicate that HSP90 regulates correct gene expression for heat acclimatization in plants. We also observed that HSP90 is involved in hormone responses in Arabidopsis. The evolutionary and functional characterizations are now being investigated.

VI. Update of The Plant Organelles Database 2 (PODB2) and release of Plant Organelles World

The Plant Organelles Database 2 (PODB2) was built to promote a comprehensive understanding of organelle dynamics. This public database is open to all researchers. PODB2 consists of five individual units: the perceptive organelles database, the organelles movie database, the organellome database, the functional analysis database, and external links. The perceptive organelles database, which was added as new content, shows organelles dynamics in response to environmental stimuli (Figure 4). The organelles movie database contains time-lapse images and 3D structure rotations. The organellome database is a compilation of static image data of various tissues of several plant species at different developmental stages. The functional analysis database is a collection of protocols for plant organelle research. The amount of included data is increasing day by day. We will add new content, which is dedicated to ultrastructures of organelles by electron microscopy, soon. It is expected that PODB2 will contribute to systems biology through the combination of the included data with other ‘omics’ data and computational analyses. In addition, we updated the website, Plant Organelles World, which is based on PODB2 as an educational tool to engage members of the non-scientific community. We expect that PODB2 and Plant Organelles World will enhance the understanding of plant organelles among researchers and the general public who want to explore plant biology.

Publication List

Original papers


Original papers (E-publication ahead of print)


Review articles


Figure 4. The graphical user interface of the perceptive organelles database in PODB2 (http://podb.nibb.ac.jp/Orgenellome).