論文の内容の要旨

Studies on diversification of organelles and membrane trafficking pathways using Marchantia polymorpha

(ゼニゴケを用いたオルガネラと膜交通経路の多様化の研究)

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Membrane traffic is a fundamental system responsible for correct transport and localization of proteins, lipids, and polysaccharides in eukaryotic organisms including plants. Among key machinery components of membrane trafficking, Rab GTPases and SNARE proteins mediate tethering and fusion between transport vesicles and target membranes, respectively. Although the molecular framework is well conserved in eukaryotic lineages, it is also known that each eukaryotic lineage has acquired lineage-specific membrane trafficking pathways during evolution, which should be involved in lineage-specific biological functions. The diversification of membrane trafficking is considered to result from, at least partly, functional differentiation of the machinery components such as Rab GTPases and SNARE proteins. However, its detailed mechanisms remain almost unknown. In this study, I aimed to unveil how membrane trafficking pathways have been diversified during land plant evolution using the liverwort, *M. polymorpha*, which is an emerging model of basal land plants, with a special interest in SNARE molecules.

I identified 34 genes for SNARE proteins in *M. polymorpha* based on the genome and transcriptome information. I then examined subcellular localization of the majority of these SNARE molecules by expressing fluorescently tagged proteins in *M. polymorpha* thallus tissues. The results obtained and comparison with the subcellular localization of orthologous products in *Arabidopsis thaliana* indicated that the membrane trafficking system has increased its complexity during land plant evolution. Through this analysis, I also succeeded in establishing reliable organelle markers in *M. polymorpha* (Chapter III).

I then carried out detailed analyses for the SYP1 group, which is remarkably expanded in seed plants, in *M. polymorpha*. I found that one of four SYP1 members in *M. polymorpha* plays an essential role in cell plate formation during cytokinesis, while its close relative is specifically expressed in oil body cells and localized to the membrane of the oil body, an organelle unique to liverworts. Observation of various organelle marker proteins and vacuolar and secretory cargos in dividing and oil body cells indicated that targeting to these organelles is accomplished by transient redirection of the secretory pathway. Furthermore, I found that none of the known organelle markers are localized to the oil body membrane, although previous studies proposed several possible organelles as origins of this liverwort-specific compartment. These results indicated that functional diversification of SYP1 members accompanied with transient alteration of transport destinations should contribute to the acquisition of new organelles in the plant lineage (Chapter IV).

For insights into molecular mechanisms of biogenesis of the oil body in *M*. *polymorpha*, I then conducted forward genetic screening for mutant plants with altered oil body morphology or distribution patterns. I have successfully isolated several putative mutants from 16,000 T-DNA-tagged lines, which will be useful to unravel how and why liverworts attained the oil body during evolution (Chapter V).