学 位 論 文 内 容 の 要 旨

博士の専攻分野の名称　博士（生命科学）　氏　名　神野 智世

学 位 論 文 題 名

Investigation of Novel Factors of Plasmodesmata Formation That Is Crucial for Multicellularity in Land Plants
（陸上植物の多細胞化の鍵を握る原形質連絡を作り出す
新奇因子の発見とその分子機構の解明）

Cell-to-cell communication is essential for the maintenance of the complex multicellular body plan of eukaryotes. Land plants have plasmodesmata (PD) as a tool for cell-to-cell communication that penetrate the cross wall between adjacent cells and connect the cytoplasm between them. The acquisition of PD is thought to have played an important role in the multicellularity of land plants. Among a general hypothesis of PD formation based on electron microscopy observations, PD are thought to be formed around strands of Endoplasmic reticulum networks that are trapped during the deposition of cell plate materials at the phragmoplast during cytokinesis. However, the molecular mechanism of PD formation remains unknown.

To explore the relationship between the multicellularity and cell-to-cell communication in land plants, I have focused on the simple structure of the protonemal tissue in the moss *Physcomitrium patens* composed of cells arranged in a single row. Protonemal cells produce stress-resistant cells called brood cells, which are induced by environmental stresses and abscisic acid (ABA), a plant hormone working for various stress responses. Brood cells are thought to function as dispersers for vegetative propagation, characterized by their rounded cell shape, thick cross walls, and easily separable cells. I first investigated changes in the multicellular body plan of the protonemal cells during brood cell induction. I found that ABA suppresses apical cell elongation and cell polarity of the cytoskeleton and vacuole in protonemal cells. I also found that ABA delays cell cycle progression in protonemal apical cells and that the protein kinase SUCROSE NONFERMENTING 1-RELATED PROTEIN KINASE 2, a key component of the ABA signaling pathway, is required for growth inhibition by ABA.

Since brood cells tend to lose their multicellularity, I hypothesized that cell-to-cell communication is lost in brood cells. Therefore, I observed PD in cross walls of brood cells by electron microscopy and found that PD density was reduced in the cross walls of brood cells. Among quantification of the density of aniline blue fluorescence signal which can stain callose at PD, I also found that ABA core module and the transcription factor ABA-INSENSITIVE 5 are important for the regulation of PD.

In the process of my research, I also hypothesized that cell-to-cell communication and the multicellular body plan are closely related. Therefore, I thought that gene mutants with cells tend to separate from each other and lose their multicellularity, as in brood cells, have less PD. From this point of view, I focused on mutants of a small GTPase, Rho of plants (ROP), and its related factors that have lost multicellularity. I hypothesized that these mutants might also have decreased PD density. Therefore, I investigated PD density in ROP knockout mutants or in knockdown mutants of the ROP-related gene that regulates the activation and localization of ROP, and found that PD density was significantly reduced in all mutants. In conclusion, I found that ROP and its regulatory factors are important factors for PD formation. Furthermore, since ROP is known to regulate actin of the cytoskeleton, I investigated mutants of Formin, which is important for actin organization, and found that PD density was also decreased in the Formin mutants. Based on these findings, I found that actin regulation by ROP and Formin, is important for PD formation. Thus, my dissertation reported the identification of several key factors important for PD formation in land plants, which were not known before. These results provide important evidence and new hypotheses for understanding the molecular mechanisms that enabled cell-to-cell communication during the evolution of plants from a unicellular to a multicellular organism.