LABORATORY OF PLANT ORGAN DEVELOPMENT

Director-General
OKADA, Kiyotaka

Postdoctoral Fellow TSUCHIDA, Yuhei
Technical Assistant HARA, Reiko
Secretary SAKAGAMI, Mari

Assistant Professor (Individual Research) TANAHASHI, Takako

Plant organs - leaves, flowers, and roots - show impressive, symmetrical shapes, based on an ordered arrangement of differentiated cells. The organs are formed from a group of undifferentiated cells located at the tip of the stem or the root. In the case of leaves, the process of organogenesis starts with the formation of a leaf primordium in the peripheral zone of the shoot apical meristem at the fixed position, following an order called phyllotaxis. Cells in the primordium then proliferate and differentiate according to three spatially fixed axes: the apical-basal axis, the lateral axis (central-marginal axis), and the adaxial-abaxial (foreside-backside) axis. In the course of proliferation and differentiation, the plant cells are believed to exchange information with neighboring or separated cells in order to regulate the organ architecture. We are trying to understand the mechanism of the information exchange between plant cells during the development of lateral organs, such as leaves, sepals, petals, stamens and carpels.

Ⅰ. Genetic approach

Recent studies of *Arabidopsis* mutant show a couple of genes are involved in the axes-dependent control of lateral organ development. FILAMENTOUS FLOWER (FIL) and YABBY3, members of the YABBY/FIL gene family encoding a protein with a zinc finger and an HMG-related domains, are among them and control the specification of the abaxial side of lateral organs. FIL gene expression was restricted at the abaxial side of the lateral organ primordia (Figure 1). On the contrary, PHABULOSA (PHB) gene encoding a class III homeobox-ZIP protein, and its homologs, REVOLUTA (REV) and PHAVOLUTA (PHV) are responsible for formation of the adaxial-side tissue. We showed that PHB is expressed in cells of the adaxial side and separated clearly from the abaxial side-specific FIL gene expressing cells, by action of microRNA165/166 which targeted the PHB, REV, and PHV messenger RNAs. A series of section analyses showed that the separation of the adaxial side- and the abaxial side-specific genes was observed in the leaf primordium of stage 0, indicating that the microRNA action is working in the earliest stage of leaf development.

Several lines of evidence showed that the fixing of the adaxial-abaxial boundary in the leaf primordium is a key step of the subsequent expression of the region-specific genes and cell proliferation and differentiation. We then screened mutants with altered adaxial-abaxial boundary position by mutagenizing seeds of transgenic Arabidopsis carrying the abaxial-side specific FIL gene promoter::GFP. One of the mutants, enf1-1, showed a fluctuating boundary position, while another mutant, enf2-1, revealed a shift of the boundary to the adaxial side. Further analysis of the mutants will unveil the molecular mechanism for determining the position of the boundary, an important process of lateral organ development.

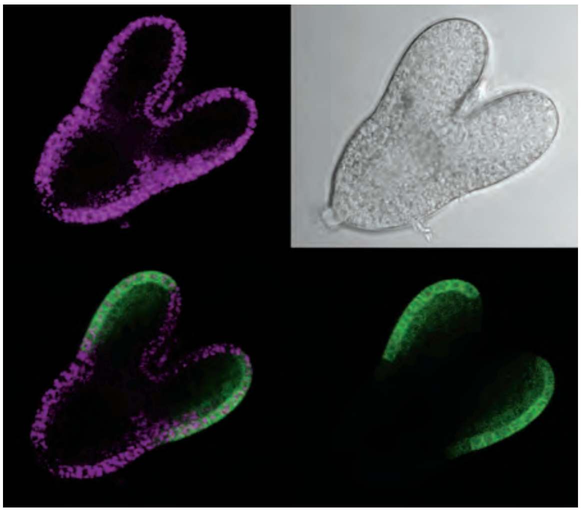

Figure 1. Region-specific expression of the FIL gene at the abaxial side-region of the developing cotyledons of Arabidopsis at the late heart stage embryo. Top right: light-field microscope picture. Top left: Pink signal show auto-fluorescence of chloroplasts. Bottom right: Green signals show the FIL promoter-drived green fluorescence protein (GFP). Bottom left: Merged picture of chloroplast auto-fluorescence and of GFP.

Ⅱ. Biochemcal approach

We are taking another approach to studying the intercellular signaling system by analyzing small peptides as candidates for intercellular signaling ligands, which are present in the apoplastic region of the shoot apical meristem. Small peptides were identified by LC/MS in the apoplastic liquid of edible cauliflower, which is a huge clump of inflorescence and premature flowers, or of *cauliflower* mutant of Arabidopsis, which makes clumped meristem identical to the vegetable. More than 400 peptide species were identified and their functions in lateral organ development are now being investigated.

Publication List

〔Original papers〕

● Ishida, T., Hattori, S., Sano, R., Inoue, K., Shirano, Y., Hayashi, H., Shibata, D., Sato, S., Kato, T., Tabata, S., Okada, K., and Wada, T. (2007). Arabidopsis TRANSPARENT TESTA GLABRA2 is directly regulated by R2R3 MYB transcription factors and is involved in regulation of GLABRA2 transcription in epidermal differentiation. The Plant Cell *19*, 2531-2543.
● Tominaga, R., Okada, K., and Wada, T. (2007). Functional analysis of epidermal-specific Myb genesgenes CAPRICE and WEREWOLF in Arabidopsis. The Plant Cell *19*, 2264-2277.