博士論文の要約

氏　　名： Tezuka, Takumi

論文題目：A study on the mechanism of dorsoventral axis formation in rice embryo

Monocots and dicots are the two major groups of angiosperms. The number of cotyledons produced during embryogenesis is one of the well-recognized differences between them. In a dicot embryo, two cotyledons are arranged symmetrically at the opposite flanks of the shoot apical meristem. On the other hand, in a monocot embryo, the shoot apical meristem is formed at the axil of one cotyledon. In grasses, the largest family in monocots, the scutellum, an organ equivalent to a cotyledon or a part of a cotyledon in monocots, absorbs nutrients from the endosperm to feed embryo. This embryo-endosperm communication is pertinent for embryo development and seedling growth of monocots and becomes possible by the arrangement of the embryonic organs where scutellum locates adjacent to endosperm and shoot faces to the outside of the seed. Hereafter, I refer to the side of the shoot in embryo as the ventral side, while the side of scutellum in the embryo is the dorsal side.

The dorsoventral axis is a unique body axis in monocot embryos and is one of the novel characters acquired when monocots are diverged from dicots. Therefore, studies on the mechanism of dorsoventral axis formation in monocot embryos are expected to give insights into the evolution of monocotyledonous plants. In my thesis, I analyzed the function of rice *BABY BOOM* genes (*BBM*s) in embryogenesis to elucidate the mechanism of dorsoventral axis formation.

bbm triple mutant embryos (hereafter called *bbm* embryos) showed morphological abnormalities associated with irregular dorsoventral axis formation. I conducted transcriptomic and *in situ* hybridization analyses and showed that the mutant embryos exhibit abnormal expressions of dorsal and ventral specific genes. Furthermore, the

expression levels of several *PIN* genes were reduced in the mutant embryos compared to those of normal ones. PIN is known to be involved in the polar auxin transport. In addition, *in situ* hybridization analysis revealed that the localized expressions of *PIN*s were lost in the mutant embryos. Therefore, I hypothesized that, in the *bbm* embryos, auxin localization and response became abnormal. I tested this using an auxin response reporter. I showed that, in the wild-type embryo, a single auxin response focus is observed at the apical tip corresponding to a part of future scutellum in the embryo, while in the *bbm* embryos, auxin response becomes irregular and unstable. Thus, it is suggested that properly localized *PIN* expressions mediated by *BBMs* followed by the formation of the auxin response focus are associated with the correct organization of the dorsoventral axis. Considering the ubiquitous expression of *BBM*s in the embryo and localized *PIN2* expression in the dorsal side, a cue for the dorsoventral axis formation must exist other than *BBMs*.

I hypothesized that auxin or other substances supplied from the endosperm, which is in contact with the dorsal side of the embryo, could be a cue to form the dorsoventral axis in the embryo. I analyzed auxin content in the embryos of *endospermless1* (*enl1*) mutants, which lack endosperm in the seed, and checked dorsoventral axis formation in *enl1* mutants. I found that auxin contents in *enl1* embryos were greatly reduced compared to the normal embryos. Notably, I did not find any change in the gene expressions of auxin biosynthesis and catabolism-related genes between *enl1* and normal embryos. Thus, it is likely that the reduction of auxin content in *enl1* is not due to defects of auxin metabolism in *enl1* mutant embryos, rather a part of auxin amount in embryo depends on development of endosperm. The expression patterns of ventral and dorsal marker genes were abnormal in *enl1* embryos. These results indicate that auxin or its precursor derived from the endosperm is required for the formation of the dorsoventral axis in the embryo.

Overall, I propose a model that the embryo-endosperm communication mediated by

auxin or its precursor and regulation of localized expression of *PINs* by *BBMs* operates to establish the dorsoventral axis in rice embryos.