

学位論文要旨
Dissertation Abstract

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学位論文題目 : Studies on expression pattern of makorin RING zinc finger protein encoding gene (*MKRN*) during embryonic and post-embryonic organogenesis in rice (*Oryza sativa* L. var. Nipponbare)
Title of Dissertation (イネ(*Oryza sativa* L. var. Nipponbare)の胚形成および発芽後器官形成におけるmakorin RING zinc finger タンパク質をコードする遺伝子 (*MKRN*) の発現に関する研究)

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Rice *MKRN* is a member of the makorin RING finger protein gene (*MKRN*) family, which encodes a protein with a characteristic array of zinc-finger motifs conserved in various eukaryotes. In the present study, we used non-radioactive in situ hybridization technique to investigate the spatio-temporal gene expression pattern of rice *MKRN* during various growth stages of rice (*Oryza sativa* L. var. Nipponbare). These growth stages broadly represented the important life events of rice, such as study at 0-1 to 2-3 days after pollination (DAP) represented the early ovule development. Also, the embryonic development was analyzed at 5-6, 8-9, 11-12, 15-16 and 45 DAP. The impact of imbibition on *MKRN* expression was studied after 1 and 6 day of imbibition, whereas early root development was observed 4 and 9 days after germination. The post-embryonic organogenesis such as lateral and crown root development was studied for *MKRN* expression at 4, 7 and 9 days after germination. The studies of these developmental stages are crucial for the further improvement in grain yield and drought resistance in rice.

During ovule development, the changes of *MKRN* expression pattern in cell sap conducting vessels of the ovule were observed. During early embryogenesis in rice, the apical-basal axis is established at 4-5 DAP. *MKRN* expression was ubiquitous during organogenesis of SAM, stele, coleoptile, coleorhiza, columella, epiblast, scutellum and

the scutellum vascular trace but not in the epidermis and lateral root cap cells in the embryo at 5-6 DAP. During the successive stages of embryonic growth such as 8-9 DAP, 11-12 DAP and 45 DAP, *MKRN* expression was gradually decreased in the differentiating cells including the coleorhiza, epiblast and the scutellum, whereas it was obvious in the SAM, RAM, provascular tissues; however it was increased again in the differentiating cells during imbibition. According to the positional distance of the tissues from the root tip, *MKRN* expression was variable in the cortex, endodermis, pericycle and the provascular tissues. The sections close to the root tip showed stronger *MKRN* expression than the sections distant from the root tip. Tissue-specific and position dependent *MKRN* expression was found during embryonic and post-embryonic root and shoot development.

During post-germination root development, *MKRN* expression was spatially similar to the embryonic root development, however in fully matured root, *MKRN* expression was localized only in the active companion cells of the phloem and it was absent in all other root cells. Also, *MKRN* was expressed in the various developmental stages of lateral root primordia, initiating with the periclinal divisions of the presumptive pericycle founder cells to its emergence. Interestingly, expression of *MKRN* was observed in the three endodermis cells adjacent to presumptive pericycle founder cells and continued in three consecutive rows of cortex parenchyma cells up to the sclerenchyma, suggesting a role of *MKRN* in the mechanism of lateral root primordia emergence. Interestingly, *MKRN* expression pattern was similar during the development of LRP and CRP; however during their emergence it was different.

During the vascular pattern development in the embryo, expression of *MKRN* was concentrated in the developing provascular tissues and SAM whereas; it was gradually reduced in the surrounding mesophyll cells. *MKRN* expression was further increased in the provascular bundle cells of 1 day imbibed embryo compared to the dry mature embryo. A similar expression pattern was also found in the coleoptile collected from the 6 days imbibed seeds. The *MKRN* expression pattern during development in various growth stages suggests an important role of makorin RING *finger* protein gene in embryonic and post-embryonic organogenesis in both apical-basal and radial developmental axes of rice. This study would assist to improve the quantitative and qualitative aspects of rice production to assure world food supply.