

Ternary and Quaternary MADS Protein Complexes Involved in Floral Organ Development in Rice**Ja-Myoung Lee, Ji-Im Park, Hee-Yeon Park, Gynheung An¹ and Yong-Hwan Moon**

Department of Molecular Biology, Pusan National University, Busan 609-735, Korea

¹Division of Molecular and Life Sciences, Pohang University of Science and Technology, Pohang 790-784, Korea

MADS genes play crucial roles in the regulation of floral organ development in plant. The function of these genes in floral organ development can be explained well with ABCDE model in Arabidopsis. Recent reports suggest that each floral organ is specified by a multimeric MADS protein complex with a specific combination of ABCDE class proteins. In particular, E class proteins, SEP proteins in Arabidopsis, are an essential component in almost all multimeric MADS protein complex identified so far. The multimeric MADS protein complexes have been well elucidated in a few dicot plants including Arabidopsis, Antirrhinum, and petunia, but in monocot the MADS protein complexes have not been reported and even functional E class proteins are not clear, yet.

In this study, to identify multimeric MADS protein complexes in rice, a representative species of monocot, we have investigated MADS proteins interacting with rice B class heterodimer, OsMADS16-OsMADS4, using yeast three-hybrid system. First, among twelve rice MADS proteins investigated, OsMADS6, OsMADS7 and OsMADS8 interacted with the OsMADS16-OsMADS4 heterodimer, suggesting that the OsMADS6, OsMADS7 and OsMADS8 may function in the form of ternary complexes including the B class heterodimer. From yeast three-hybrid screen using cDNA libraries of young panicles and the OsMADS16-OsMADS4 heterodimer as bait, OsMADS6 and OsMADS7 were isolated, confirming previous results, the OsMADS16-OsMADS4-OsMADS6 and OsMADS16-OsMADS4-OsMADS7 ternary complexes. The OsMADS6 is classified into AGL6 subfamily, and the OsMADS7 and OsMADS8 are classified into AGL2 subfamily, indicating that the B class heterodimer interacts with two groups of MADS proteins. In particular, the very high levels of amino acid sequence similarity that OsMADS7/OsMADS8 share with Arabidopsis SEP proteins, together with almost identical expression pattern, suggests that E class proteins in rice like those in dicot may play an essential role in the formation of MADS protein complexes.

On the other hand, we have investigated domains of OsMADS4 responsible for the formation of the OsMADS16-OsMADS4-OsMADS6 and OsMADS16-OsMADS4-OsMADS7 ternary complexes using yeast three-hybrid system. As results, we found that both K and C domains are responsible for the formation of the ternary

MADS protein complexes, and that especially the region including whole K and C domains is necessary for the interaction, indicating that C domain as well as K domain is involved in the formation of multimeric MADS protein complexes.

Furthermore, to investigate OsMADS proteins which are interacting with OsMADS16-OsMADS4-OsMADS6 complex, we performed GAL4-based yeast four-hybrid experiment. As results, it was revealed that OsMADS6, OsMADS7, and OsMADS17 strongly interact with the OsMADS16-OsMADS4-OsMADS6 tertiary complex.

Taken together, our results suggest that multimeric rice MADS protein complexes may play an important role in the specification of floral organ development probably with fine regulation, and that the MADS protein complexes may be conserved between monocot and dicot.