

Chareerat Mongkolsiriwatana 2008: Expression Analysis of Photoperiod Responsive Genes in Rice (*Oryza sativa* L.) KDML 105. Doctor of Philosophy (Genetics), Major Field: Genetics, Department of Genetics. Thesis Advisor: Associate Professor Surin Peyachoknagul, Dr.Agr. 108 pages.

GeneChip DNA microarray was used to identify photoperiod responsive genes of rice (*Oryza sativa* L.) KDML 105. A comparative analysis of gene expression changes between short day (SD) and long day (LD) conditions was performed with three independent biological replications. A total of 184 probe sets were selected as differentially expressed genes on the basis of their expression ratios with t-test p value (fold changes  $\geq 2$ ,  $p \leq 0.05$ ) and were verified with RT-PCR and real-time PCR. Among them, 79 genes were up-regulated while 105 genes were down-regulated. Those genes were classified into nine classes from their putative functions, i.e., unknown, transcription factor, defense/stress, metabolism, growth/structure, processing, signaling, transport and energy transduction. Pathway analysis revealed that the photoperiod response is involved in photosynthesis, carbohydrate metabolism, circadian clock, phytochrome signaling, hormone signaling and miRNA synthesis pathways. Several flowering time related genes were also associated with those pathways; the floral inducers were up-regulated while the floral repressors were down-regulated, including the targets of miRNAs such as *AP2* floral repressor. Expression analysis of miRNA of *AP2*, *miRNA172a*, showed that its expression was induced by SD light. This indicated that *miRNA172a* was involved in the regulation of photoperiodic flowering time in rice via the down-regulation of *AP2*. Monitoring expression of photoperiodic flowering time genes during SD induction revealed that floral inducers, i.e., *Hd3a* and *API like* were induced in day 6 and day 10, respectively, while floral repressor, *AP2* was suppressed in day 4. This showed that *API like* was a downstream of *Hd3a* and their expression were probably induced by the down-regulation of *AP2*.

To elucidate the regulatory mechanism of photoperiod response, the *cis*-regulatory elements of photoperiod responsive promoters were analyzed. The results showed that the transcription of photoperiod responsive genes is controlled by light in coordination with hormones and stress responses. GARE motif and G-box are the coordinated motifs integrating gibberellins to photoperiod while MBS and G-box are the coordinated motifs integrating ethylene or abscisic acid and stresses to photoperiod. Phytochrome A (phyA) regulated genes were further identified using the specific organization of *cis*-regulatory elements. The data showed that phyA is involved in the transcriptional regulation of flowering time genes either by activation of floral inducers or suppression of floral repressors. So far, two novel *cis*-regulatory elements which are specific to daylength were identified. The novel A-rich element is specific to LD light involved in the regulation of phyA and circadian rhythm to inhibit flowering whereas the novel GC element is specific to SD light involved in the regulation of gibberellins signaling to promote flowering. Taken altogether, the photoperiodic flowering pathway of KDML 105 was proposed. The flowering transition is controlled by phyA and circadian rhythm in coordination with hormone signaling, stress responses and metabolic state. The floral development is switched by down-regulated *AP2* by *miRNA172a* and up-regulated *Hd3a* leading to activation of floral meristem identity gene, *API like*, *CAL* and *OsMADS1*, via the meristem maintenance CLAVATA pathway.

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