



Identification of genes associated with the increased number of four-seed pods in soybean (*Glycine max* L.) using transcriptome analysis

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Genet. Mol. Res. 14 (4): 18895-18912 (2015)

Received August 16, 2015

Accepted October 9, 2015

Published December 28, 2015

DOI <http://dx.doi.org/10.4238/2015.December.28.39>

ABSTRACT. Seed number per pod is an important component of yield traits in soybean (*Glycine max* L.). In 2010, we identified a natural mutant with an increased number of four-seed pods from a soybean variety named 'Jinong 18' (JN18). Subsequent observations indicated that the trait was stably inherited. To identify and understand the function of genes associated with this mutant trait, we analyzed the genetic differences between the mutant (JN18MT01) and source variety (JN18) by transcriptome sequencing. Three types of tissues, axillary buds, unfertilized ovaries, and young pods at three different growth stages, V6, R1, and R3, were analyzed, respectively. The sequencing results yielded 55,582 expressed genes and 4183 differentially expressed genes (DEGs). Among these, the log₂ ratio value of 162 DEGs was >10, and 13 DEGs had overlapping expression at three different growth stages. Comparisons of DEGs among three different growth stages yielded similar results in terms of the percentage of genes classified into each gene ontology (GO) category. DEGs were classified into 25 different functional groups in clusters of orthologous groups analysis. Proportions of the main functional genes differed significantly over developmental stages.

A comparison of enriched pathways among the three developmental stages revealed that 646 unigenes were involved in 103 metabolic pathways. These results show that the development of four-seed pods is associated with a complex network involving multiple physiological and metabolic pathways. This study lays the foundation for further research on cloning and on the molecular regulation of genes related to the four-seed pod mutation.

Key words: Soybean; Four-seed pod mutant; Transcriptome analysis; High-throughput Illumina sequencing; Developmental stages