

Genome-wide analysis and expression profiling of half-size ABC protein subgroup G in rice in response to abiotic stress and phytohormone treatments

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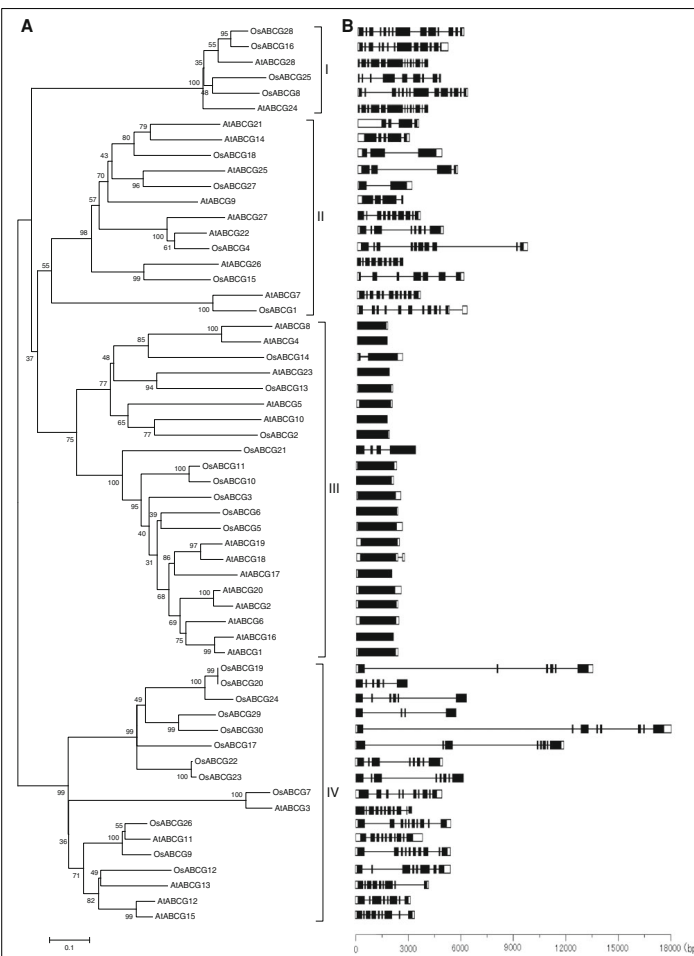
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Abstract

The roles of the proteins encoded by half-size adenosine triphosphate-binding cassette transporter subgroup G (*ABCG*) genes in abiotic stress responses are starting to be established in the dicot model *Arabidopsis thaliana*. In the monocot model rice, the functions of most half-size *ABCG* proteins in abiotic stress responses are unknown. *Rcn1/OsABCG5* is an essential transporter for growth and development under abiotic stress, but its molecular function remains largely unclear. Here, we present a comprehensive overview of all 30 half-size *ABCG* genes in rice, including their gene structures, phylogeny, chromosome locations and conserved motifs. Phylogenetic analysis revealed that the half-size *OsaBCG* proteins were divided to four classes. All seven rice intronless genes, including *Rcn1/OsABCG5*, were in Class III, like the 12 intronless *ABCG* genes of *Arabidopsis*. The EST and FL-cDNA databases provided expression information for 25 *OsaBCG* genes. Semi-quantitative and quantitative RT-PCR analyses demonstrated that seven *OsaBCG* genes were up-regulated in seedlings, shoots or roots following treatments with abiotic stresses (6°C, 17°C, 42°C, NaCl, or mannitol) and abscisic acid. Another 15 *OsaBCG* genes were up-regulated under at least one of the abiotic stress conditions and other phytohormones besides abscisic acid. Hierarchical clustering analysis of gene expression profiles showed that expression of the *OsaBCG* genes could be classified into four clusters. The *Rcn1/OsABCG5* cluster was up-regulated by abscisic acid and included *OsaBCG2*, 3, 13 and 27. The present study will provide a useful reference for further functional analysis of the *ABCG*s in monocots.

Results

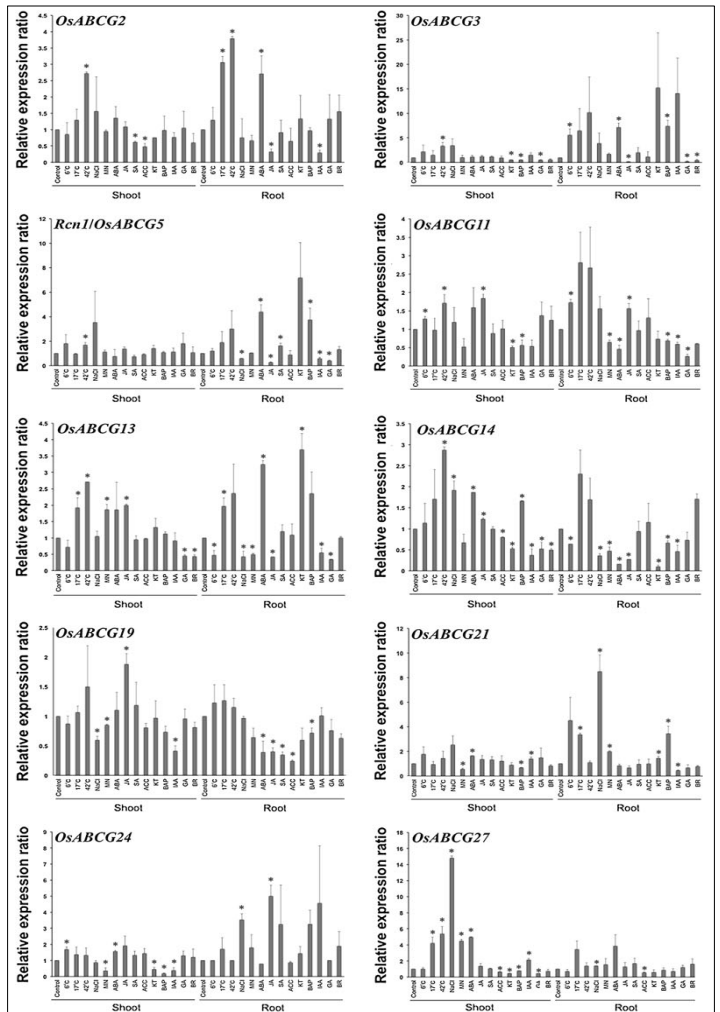
1. Phylogenetic analysis of half size *ABCG* proteins.



Phylogenetic tree of rice and *Arabidopsis* half-size *ABCG* proteins (a). Full-length sequences of 30 *ABCG* proteins of rice were compared with corresponding sequences for 28 members of *Arabidopsis* *ABCG* proteins to construct a tree with Mega4 software using the Neighbor-Joining (NJ) method (5,000 replicates). Exon-intron organization of corresponding *ABCG* genes (b). Exons and introns are represented by black boxes and lines, respectively. Open boxes indicate untranslated regions

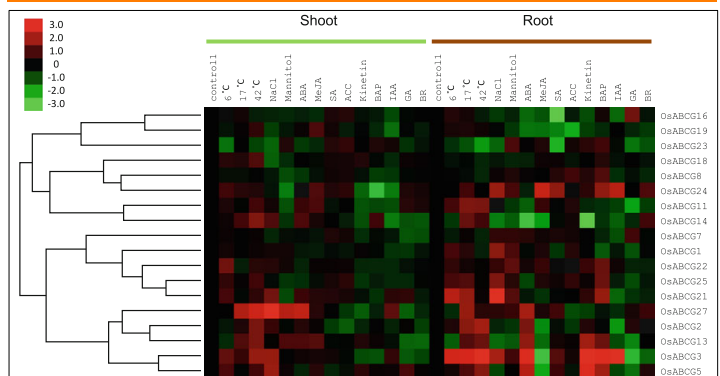
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2. Expression of part of the rice *ABCG* protein genes



Expression profiles of *OsaBCG2*, *OsaBCG3*, *Rcn1/OsABCG5*, *OsaBCG10*, *OsaBCG11*, *OsaBCG13*, *OsaBCG14*, *OsaBCG19*, *OsaBCG24*, and *OsaBCG27* as determined by quantitative RT-PCR using total RNA from shoot and root sampled 3 h after treatments. Relative amount of amplified product was normalized against *OsUBQ10* transcripts. Each value represents the average of two or more biologic replicate experiments in which each value is the mean of three separate quantitative RT-PCR reactions (\pm SE). Asterisks indicate significant differences ($p < 0.05$) compared with control

3. Hierarchical clustering analysis of rice *ABCG* genes



Hierarchical cluster of expression profiles of rice half-size *ABCG* genes. *OsaBCG* genes were considered to be expressed if the normalized intensity value exceeded 0. Expression levels are illustrated by graded color scale, representing relative expression levels of -3, -2, -1, 0, 1, 2, and 3. Red, green, and black represent positive, negative, and zero, respectively