

RNA-Seq and validation analysis on the important genes involved in early responses to salinity stress of Malaysian rice seedlings (*Oryza sativa* ssp. *Indica*)

ABSTRACT

Salinization of rice cultivation land has progressively enlarged, thus negatively impaired the world's rice bowl. Due to the polygenic nature and complexity of salinity tolerance mechanisms in plants, the development of new rice varieties with better adaptation to salinity has become a great challenge. Regarding this, transcriptomic profiling has been seen as a promising approach for a holistic understanding of salinity tolerance mechanisms in rice. Using Illumina RNA-Seq method, transcriptomes of two contrasting Malaysian rice varieties named as salt-tolerant MR211 and salt-sensitive MR220 were analysed within a short-term exposure (9 h) to salt-shock treatment (12 dS m⁻¹) at early seedling stage. Transcriptomic analysis using Tuxedo package enabled the identification of 252 differentially expressed genes (DEGs). Interestingly, 93.3% of the DEGs (n=235) were identified as higher and specifically expressed in salt-tolerant MR211 compared to the sensitive variety (MR220's FPKM \leq 0). This group of DEGs was assigned in 33 KEGG pathways, with the highest number of transcripts accounted in purine and thiamine metabolism pathways. Meanwhile, functional annotation analyses revealed the presence of regulatory genes, annotated functional and unknown genes involved in various salt adaptation mechanisms in the salt-tolerant variety MR211. The expression accuracy and reproducibility of the 252 DEGs identified from the RNA-Seq experiment were further verified through semi-quantitative RT-PCR followed by real time PCR analysis.

Keyword: Comparative transcriptomes; Salt stress; Indica rice; Seedling stage