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Functional characterization of ABA signaling components using transient gene expression in rice protoplasts

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Abstract

The core components of ABA-dependent gene expression signaling have been identified in *Arabidopsis* and rice. This signaling pathway consists of four major components; group A OsbZIPs, SAPKs, subclass A OsPP2Cs and OsPYL/RCARs in rice. These might be able to make thousands of combinations through interaction networks resulting in diverse signaling responses. We tried to characterize those gene functions using transient gene expression for rice protoplasts (TGERP) because it is instantaneous and convenient system. Firstly, in order to monitor the ABA signaling output, we developed reporter system named pRab16A-fLUC which consists of *Rab16A* promoter of rice and luciferase gene. It responses more rapidly and sensitively to ABA than pABRC3-fLUC that consists of ABRC3 of *HVA1* promoter in TGERP. We screened the reporter responses for over-expression of each signaling components from group A OsbZIPs to OsPYL/RCARs with or without ABA in TGERP. OsbZIP46 induced reporter most strongly among OsbZIPs tested in the presence of ABA. SAPKs could activate the OsbZIP46 even in the ABA independence. Subclass A OsPP2C6 and -8 almost completely inhibited the OsbZIP46 activity in the different degree through the SAPK9. Lastly, OsPYL/RCAR2 and -5 rescued the OsbZIP46 activity in the presence of SAPK9 and OsPP2C6 dependent on ABA concentration and expression level. By using TGERP, we could characterize successfully the effects of ABA dependent gene expression signaling components in rice. In conclusion, TGERP represents very useful technology to study systemic functional genomics in rice or other monocots.

Keywords: ABA, PYR/PYL/RCARs, PP2Cs, bZIP, rice

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