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Identification of the Biological Function of Rab-GGT β-Subunits by Reverse Techniques

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ABSTRACT

Protein prenylation is a post-translational process where lipids are added to carboxyl end groups of amino acids, which allows proteins to function properly in the eukaryotic cell. The job of prenylation is to target certain proteins to specific membranes and promote desirable protein-protein interactions. In our study we used reverse genetics techniques to investigate the function of protein prenylation in plant development. To discern the function of protein prenylation, we examined the phenotypic changes caused by specific gene disruptions. In this study the model organism Physcomitrella patens (moss) is utilized due to its simple structure, limited quantities of tissues and cells, fully sequenced genome, and high gene targeting efficiency.

Rab geranylgeranyl transferase-II (Rab-GGT) is one of three enzymes that can perform protein prenylation – however, the function of Rab-GGT in vivo is largely unknown. P. patens has one copy of Rab-GGT α subunit gene (PpRGTA1) and two copies of β subunit genes (PpRGTB1 and PpRGTB2). This study focuses on the role of the Rab-GGT β subunit in the P. patens which will likely translate to other eukaryotic organisms. Studies have demonstrated that the knockout of either PpRGTB1 or PpRGTB2 results in no visible phenotype, which leads us to believe that these genes are functionally redundant. Additionally, the knockout of both PpRGTB1 and PpRGTB2 genes has shown to be lethal, which indicates that Rab-GGT is required for viability. To determine the function of Rab-GGT, we implement the reverse genetics approach, RNA interference (RNAi), to down-regulate the expression level of PpRGTB2 in the PpRGTB1 knockout background to observe the phenotypic consequences of minimal Rab-GGT β subunit gene expression.

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