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The DONGLE gene encodes phospholipase A1 catalyzing JA biosynthesis and has crucial role in wounding response in Arabidopsis

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Objectives

Through molecular biological tool and genetic analysis, we tried to characterize *dongle-D*.

Materials and Methods

1. Material : *Arabidopsis thaliana* (Columbia), , Methyl Jasmonate (-Aldrich 392707) and etc
2. Method : To analyze mutant, we use various molecular biological tool including TAIL-PCR, RT-PCR, genomica southern and *in vivo* expression using protoplast system

Results and Discussion

Using activation tagging mutant screening, we had isolated a mutant that exhibited dwarfism, weak apical dominance, infertility, short petioles, and round shaped leaf morphology. Because of its round shaped leaf morphology, we have designated the mutant as *dongle-D*, dongle means round shape in Korean. In this mutant, the *At1g05800* gene was overexpressed by the CAMV 35S enhancers of T-DNA. The *At1g05800* gene encodes putative phospholipase protein that exhibited homology with jasmonic acid (JA) biosynthetic gene, *Defective in Anther Dehiscence 1 (DAD1)*. Integration of transgene which overexpresses the *At1g05800* recapitulated phenotypes of the mutant. Therefore, we have concluded that various phenotypes of the mutant were induced by the overexpression of the *At1g05800*, *DGL*. Consistent to the homology with *DAD1*, the overexpression of *DGL* induced the upregulation of JA responsive genes, including *VSP1* and Thi2.1. As well as, the JA and methyl-JA was increased in *dgl-D*. These results suggest that DGL also has a role in jasmonic acid biosynthetic pathway same as DAD1. DGL is characteristic to a putative N-terminal transit peptide and a conserved lipase domain. DGL:GFP fusion protein localized in chloroplast and DGL protein expressed in *E.coli* hydrolyzed phospholipids in an *sn*-1-specific manner. These results indicate that DGL is involved in initial step of JA to produce linolenic acid. This is supported by the fact that phenotype of *dgl-D* is suppressed when *dgl-D* is crossed with JA signaling mutant or JA synthetic mutant defective in downstream gene of *DAD1*. 11 hypothetical genes were reported to be homologous to *DAD1* in *Arabidopsis* genome. We analyzed DAD1 and 6 homologues with transit peptide and lipase domain to classify them by their function. *DAD1*, *DGL*, and *At4g16820* expressed extremely low level during seedling stage. In addition, expression of three genes by wounding showed same pattern with an increase in 30minutes. This implies that *DAD1*,*DGL*, and *At4g16820* have important role in JA synthesis to gain wounding resistance. In conclusion, DGL catalyze initial step and JA biosynthesis. Especially, DGL possibly have essential role in wound induced JA response together with *DAD1* and *At4g16820*.

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