Understanding DEVIL-like (DVL) small peptide mediated signaling in Plants

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Abstract

The aim of this project is to elucidate the signaling pathway mediated by the DEVIL-like (DVL) small peptides in plants, using Arabidopsis thaliana as a model organism due to its well-characterized genome, rapid life cycle, and suitability for genetic manipulation. DVL peptides are signaling molecules localized at the plasma membrane and their over expression causes reduction in root growth and change in leaf and fruit shape. However, how DVLs regulate these developmental processes is still unclear. To identify components of the DVL signaling pathway, a mutant library has been generated by treating seeds of the DVL overexpressing plants with ethyl methanesulfonate (EMS). We will screen for mutants that no longer show the altered phenotypes. Several putative mutants have been acquired, which might contain mutations in the receptors for the DVL peptides or downstream effectors, or the DVL gene itself. These mutants will be instrumental in our effort to identify the components of the DVL signaling cascade, which in tern will help us to understand how the DVL peptides regulate root development.

Background

DVL peptides are a conserved family of small plant-specific regulatory proteins found in land plants. These short peptides function as signaling modulators that help coordinate organ growth and developmental patterning. In plants, small peptides commonly regulate development by interacting with membrane-bound receptor-like kinases, triggering downstream signaling pathways that influence cell proliferation and differentiation. DVL peptides are thought to function within this signaling framework.

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Purpose

In this study, we aim to identify different components involved in the DVL pathway by starting with DVL overexpressing plants then screening for mutants that revert back to wild type phenotypes. These revertant lines likely carry mutations in genes required for DVL-mediated signaling. The mutations will be identified through mapping using molecular methods, allowing us to uncover genetic components that function downstream or in cooperation with DVL.

Methods

First, we created a mutant library by treating seeds overexpressing DVL with ethyl methanesulfonate (EMS) to induce mutations. We are currently in the process of sterilizing the seeds with a bleach based solution then plating them on petri dishes for initial growth. In the stage of initial growth we are looking for phenotypic variation hoping to observe a longer primary root length which would possibly indicate altered components of the DVL pathway. After selecting plants with longer primary root length they will be placed in soil and allowed to grow so other phenotypic variations in leaf and fruit shape can be seen (another possible marker for DVL pathway mutation). Finally, after selecting an organism with the desired traits we will perform DNA sequencing to try and pinpoint where in the genome the mutations took place by comparing the revertant plant with DVL overexpressed plant genome.

Relevance

By mapping mutations that alter DVL pathway activity, this study aims to uncover key receptors or downstream regulators that control root elongation and organ morphology. Defining this pathway will contribute to fundamental knowledge of plant growth coordination and may reveal novel genetic targets for modifying plant architecture in agricultural systems.

Expected Results

Overexpression of DVL peptides in Arabidopsis thaliana leads to noticeable changes in plant morphology. Plants exhibit reduced primary root length, shortened and rounder leaves, and more compact fruits. Mutant plants with disruptions in components of the DVL signaling pathway are expected to display revertant phenotypes that more closely resemble the wild-type form, despite the presence of DVL overexpression. Root length will be observed in the early stages of plant development with hopes to recover seedlings that have longer roots. Finding these mutants however, is very rare due to the small number of genes in the DVL pathway compared to the whole genome, with the redundancy factor only increasing the difficulty. After successfully obtaining a plant with the desired root morphology it will then be grown in hopes to observe a more elongated leaf and typical fruit development which will indicate possible disruption of DVL-mediated signaling.

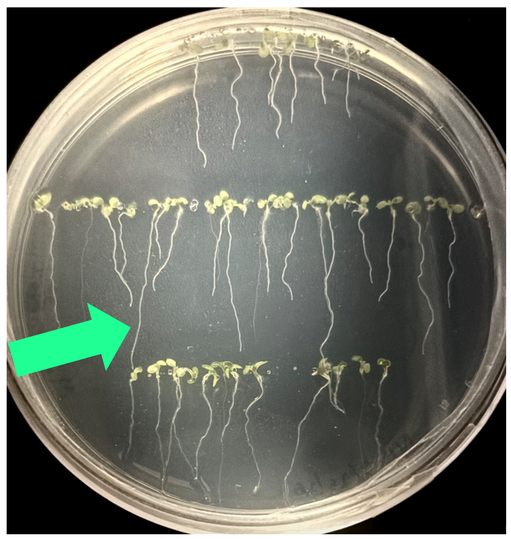

Image 1. An example of mutant screen with green arrow indicating plausible mutant

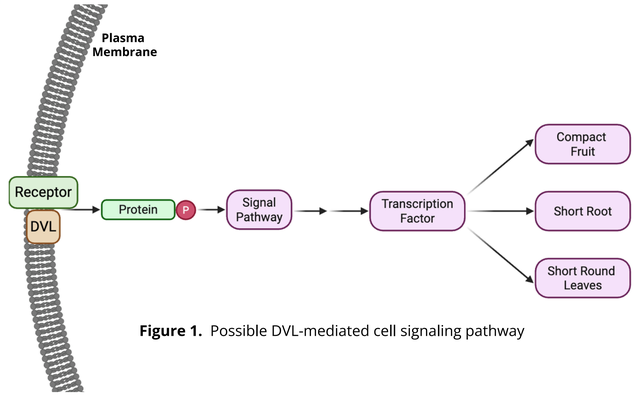

Figure 1. Possible DVL-mediated cell signaling pathway

Limitations

Our biggest limitation came during the sterilization process. We are still experiencing fungal contamination during the plating stage which prevents us from using seeds in further steps of our experiment. We are able to minimize fungus growth with increased levels of bleach; however, this also inhibits growth and germination rates. Currently, we are working on a sterilization protocol that minimizes fungus while not inhibiting growth and germination rates. Another limitation arises from the redundancy of the DVL peptides. They have an additive genetic effect, which means removing one peptide can be partially masked by the expression of the others. This created problems for identifying mutants during the screening phase of the experiment

References

Yang, L., et al. (2024) Stem-cell expressed DEVIL-like small peptides maintain root growth under abiotic stress via ABA signaling. Plant Physiology. 194: 2372–2386. DOI:10.1093/plphys/kiad659.

Guo P, Yoshimura A, Ishikawa N, et al. 2015. Comparative analysis of the RTFL peptide family on the control of plant organogenesis. J Plant Res. 128(3):497–510.