Cell Adhesion in *Arabidopsis thaliana*

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Cell adhesion is an integral process to plant growth, morphogenesis, and organism development, as without it cells would fall apart from each other. These processes of adhesion are dependent on the composition of the cell wall that surrounds all cells, and in particular, pectin is essential. By analyzing mutant cells that do not perform proper cellular adhesion, components that lead to adhesion can be identified. The lab has already isolated a family of 5 ELMO proteins that are all part of the same gene family, predicted to be in the Golgi, and each are likely to affect cell adhesion as their mutants lead to cells separating from each other. These proteins were found by analyzing mutant strains of *A. thaliana* and staining the cells with Ruthenium Red dye that binds pectin and can only penetrate and thus stain abnormal cell walls. As shown by this staining, some mutants cause stronger cell adhesion defects than others. Specifically, elmo1 and elmo4 mutants have strong phenotypic effects whereas elmo2, elmo3, and elmo5 present a phenotype remarkably similar to wild type.

This summer I worked to understand why some elmo mutants have worse phenotypic effects than others. We hypothesized that elmo2, elmo3, and elmo5 mutants without strong adhesion defects may be upregulating the other ELMO proteins to compensate for their mutation. To analyze the phenotypic differences between the five elmo mutants, we measured gene expression within each of the mutants. Using quantitative PCR, we screened for ELMO1-5 gene expression in wild type, elmo1-5 mutants, and the double mutants elmo1/2, elmo1/3, and elmo1/5. We found no upregulation of ELMO1, ELMO2, ELMO3, or ELMO5 in other elmo mutants. However, in the most extreme phenotype of elmo1/2, ELMO4 is upregulated, presumably to compensate for reduced family expression. Put together, these gene expression results indicate that the ELMO proteins are generally not compensating for one another unless the mutation is extreme, and this is only seen for ELMO4. This has left me with more questions as to what specific roles each ELMO plays, and why some are more integral to proper cellular adhesion than others.

Specifically, I have more questions regarding the structural role of the ELMO proteins. Past research has shown that the ELMO1 protein interacts closely with other pectin biosynthetic enzymes. Specifically, immunoprecipitation results pulled down ELMO1, ELMO4, QUA1, QUA2, and GAUT9 together. While the lab has hypothesized that ELMO1 may be a scaffold protein for these other pectin biosynthetic enzymes, the specific physical interactions between these proteins remain unknown. It is also unknown if ELMO acts as a scaffold or a Golgi retention protein, or both. This fall, I plan to evaluate the interactions of these proteins by focusing on characterizing the role of QUA2. Specifically, I will create a QUA2-GFP (Green Fluorescent Protein) fusion protein and use confocal microscopy to determine if in an elmo1 mutant QUA2 is retained in the Golgi or released into the cell wall. By applying the skills I have learned this summer, I hope to characterize the structural role of QUA2 as I continue to build my body of research for my senior honors thesis.

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