

GUARD CELL POLYGALACTURONASE1 regulates cell expansion and stomatal dynamics in *Arabidopsis thaliana*

Yue Rui^{1,2}, Chaowen Xiao^{1,5}, Hojae Yi³, Baris Kandemir⁴, James Z. Wang⁴, Virendra M. Puri³, and Charles T. Anderson^{1,5}

¹Department of Biology, ²Plant Biology Interdepartmental Graduate Program, ³Department of Agricultural and Biological Engineering, ⁴College of Information Sciences and Technology, ⁵Center for Lignocellulose Structure and Formation, The Pennsylvania State University, University Park, PA 16802 USA

ABSTRACT

Stomatal guard cells are dynamic cell pairs that control transpiration and gas exchange at the plant surface. Distinct from other epidermal cell walls that undergo irreversible expansion, the walls of guard cells must be both strong and elastic to withstand tremendously high turgor pressure and allow for repeatable expansion and contraction. However, genetic evidence for the importance of certain wall components, especially pectins, in stomatal function is still lacking. Using published guard cell-specific transcriptome data, we identified a panel of pectin-modifying genes that are up- or down-regulated in *Arabidopsis thaliana* guard cells, and examined stomatal development and function in homozygous T-DNA knockout mutants for these genes. We have characterized a candidate gene named *GUARD CELL POLYGALACTURONASE1 (GCPG1)*, which encodes a polygalacturonase that cleaves pectin backbones. Compared to wild-type controls, developing stomata are reduced in size in *gcpg1* knockout mutants. In abscisic acid (ABA)-induced stomatal closure assays, wild-type stomata close smoothly, whereas *gcpg1* knockout stomata exhibit more variable patterns of aperture change at both the cell population level and at the level of individual stomata. Primary root growth, etiolated hypocotyl length, and rosette size are also reduced in *gcpg1* mutants. These phenotypes are complemented by transgenic expression of *GCPG1*. When *GCPG1* is overexpressed, root elongation and rosette size are enhanced. *GCPG1* is expressed in guard cells and pavement cells that contact guard cells, and is also expressed in roots, hypocotyls, cotyledons, rosette leaves, stems, flowers, and siliques. GFP-tagged GCPG1 is localized in the apoplast and accumulates at stomatal pore initiation sites in developing guard cell pairs. Together, these data suggest that GCPG1 is essential for maintaining wall elasticity in stomatal dynamics and regulating wall expansion in plant growth. We are currently performing biochemical analyses in *gcpg1* mutants and *GCPG1* overexpression lines to investigate the molecular mechanisms underlying their cellular and growth phenotypes, which, together with a finite element-based computational model we are constructing, will provide a more comprehensive understanding of how GCPG1 modifies guard cell walls to facilitate stomatal dynamics.