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Succinoglycan Production in Agrobacterium tumefaciens

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Secretion of an acidic polysaccharide known as succinoglycan is required for plant host invasion by the bacterial phytopathogen, *Agrobacterium tumefaciens*, leading to the formation of crown galls. How is succinoglycan used during plant invasion? One possibility is that succinoglycan is involved in conferring pH tolerance in the acidic environment surrounding the roots of a plant. Recently, our group has accumulated data supporting another possibility: that succinoglycan protects bacteria from growth inhibition and cell wall damage. Upon the depletion of the essential cell wall synthase PBP1a, *A. tumefaciens* loses its ability to elongate and begins upregulating production and secretion of succinoglycan. To explore the link between cell wall synthesis and succinoglycan production, I dissected various components of the succinoglycan regulatory and biosynthesis pathways and observed changes in shape with microscopy. I found that there is indeed a link between succinoglycan biosynthesis and cell wall synthesis. Additionally, I localized the succinoglycan secretion machinery and found that it colocalizes with the growth pole, further implicating a connection between PBP1a and the regulation of succinoglycan production. In an effort to further investigate pathways regulating succinoglycan production upon loss of PBP1a activity, I developed a forward genetic approach to screen a library of transposon mutants for misregulation of succinoglycan production upon depletion of PBP1a. The screen relies on a high throughput method of detecting a dye called calcofluor white that binds succinoglycan and fluoresces under a UV light. I optimized the screen for use in a plate reader and ran 48 mutant colonies as a trial. I expect that after screening more mutants, I will identify additional proteins involved in the regulation of succinoglycan and shed more light on the link between cell wall synthesis and succinoglycan production in *A. tumefaciens*.