

Characterization of the interaction between a microbial ice nucleation protein and a plant antifreeze protein

Heather Tomalty and Virginia K. Walker

Department of Biology, Queen's University, Kingston ON

Through the production of ice nucleation proteins (INPs), certain strains of plant epiphytes are capable of encouraging the crystallization of water at high subzero temperatures. Viewed as plant pathogens, many of these ice-nucleating (INA⁺) microbes are thought to use their INPs to wound the plant, permitting access to a rich pool of nutrients within the plant. However, at low temperatures, some host plants of INA⁺ bacteria express antifreeze proteins (AFPs). These proteins adsorb to the surface of ice crystals, controlling their growth. While it is largely accepted that the function of plant AFPs is to control the growth of large ice crystals once freezing has occurred, it is also possible that plant AFPs could function as a defensive mechanism against the detrimental effects of microbial INPs. To explore this hypothesis, recombinant AFPs from the perennial ryegrass *Lolium perenne* were combined with the INP from the most prevalent INA⁺ epiphyte found associated with *L. perenne*, *Pseudomonas syringae*. Remarkably, the plant AFP was able to moderately depress the freezing point of the bacterial INP; while in contrast, a Type III AFP did not depress the freezing point, but instead appeared to modestly enhance INP activity. Ice recrystallization inhibition and ice morphology assays were also performed and INPs were not found to have any effect on AFP activity. Assays using mutated *L. perenne* AFPs will also be presented to further explore this INP-AFP interaction.

Acknowledgments: Scholarships to HT were provided by OGS and NSERC CGS, with the support of an NSERC (Canada) grant to VW.