Encapsulation of Biologicals for Agriculture: Eco-Friendly Solutions for a Sustainable Future

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Background/Objectives. Rising temperatures due to global warming create stressful environments for crop fields and cause a reduction in crop yield, threatening food security. Moreover, climate change alters the ecosystem, generating new niches for insects and posing significant challenges for the farmer. The extensive use of chemical pesticides to treat these new pests will increase the pollution burden faced by downstream communities as flooding intensifies, not to mention the large amount of fossil fuels required to generate the billions of pounds of pesticide currently needed annually, further propagating the effects of climate change. So, what is the solution to stop this vicious cycle? - Biologicals. Biologicals such as proteins, nucleic acids, and whole-cells require minimal energy input for growth and production, reducing the carbon footprint compared to traditional chemical syntheses. Additionally, beneficial microbes can be used to stimulate plant growth under stressful conditions created by global warming, while nucleic acids can be guickly reconfigured to target insects with high specificity and no off-target responses, which is imperative in this fast-changing environment induced by climate change. Unfortunately, biologicals are significantly less stable than chemicals, so shelf-stability and field-longevity are real challenges in the development of biologicals in agriculture. Here, we describe two eco-friendly concepts started at Battelle to improve the stability of biologicals for crop stimulation and crop protection.

Approach/Activities. Battelle is developing two different encapsulation strategies for biologicals: the Superhydrophobic Water-Resistant EncapsuLation (SWEL) technology to stabilize plant growth-promoting bacteria for biological seed treatment, and a technology in early stages of development for the encapsulation of nucleic acids for use as biopesticides. The SWEL formulation increases hydrophobicity via inclusion of soybean lipids that render capsules impermeable to water or aqueous solutions, places microbial growth in stasis with improved viability, and prevents desiccation. This process requires low energy input, and it uses cheap starting materials. On the other hand, we are developing a novel encapsulation approach to stabilize small-interfering RNA (siRNA), a molecule that has garnered significant attention in the agriculture industry in recent years. Our approach to encapsulate siRNA exploits a biologically generated molecule as the encapsulant, so its production results in less energy output compared to synthetic polymers.

Results/Lessons Learned. We used fluorescent microscopy to validate encapsulation of growth-stimulating bacteria *Pseudomonas protogens* in SWEL capsules and demonstrated improved stability on-shelf and on-seed of both *Pseudomonas protogens* and *Bradyrhizobium japonicum* compared to native microbes. Importantly, SWEL capsules open and release microorganisms at the time of seed germination, which was confirmed by development of nodules with longer shoot and root length which is correlated with N-fixation during later phase of plant development. While the SWEL technology proof-of-concept is established, the siRNA encapsulation technology is less mature, but we have some preliminary results that show promise. Our biological-based framework can encapsulate nearly 100% of the siRNA after four hours. We are working to improve siRNA loading efficiency by engineering the scaffold, and plan to pursue stability tests in the near future. We believe these two eco-friendly encapsulation

approaches will accelerate the use of biologicals in agriculture, providing solutions for farmers to combat the changing climate while maintaining a reduced carbon footprint.