

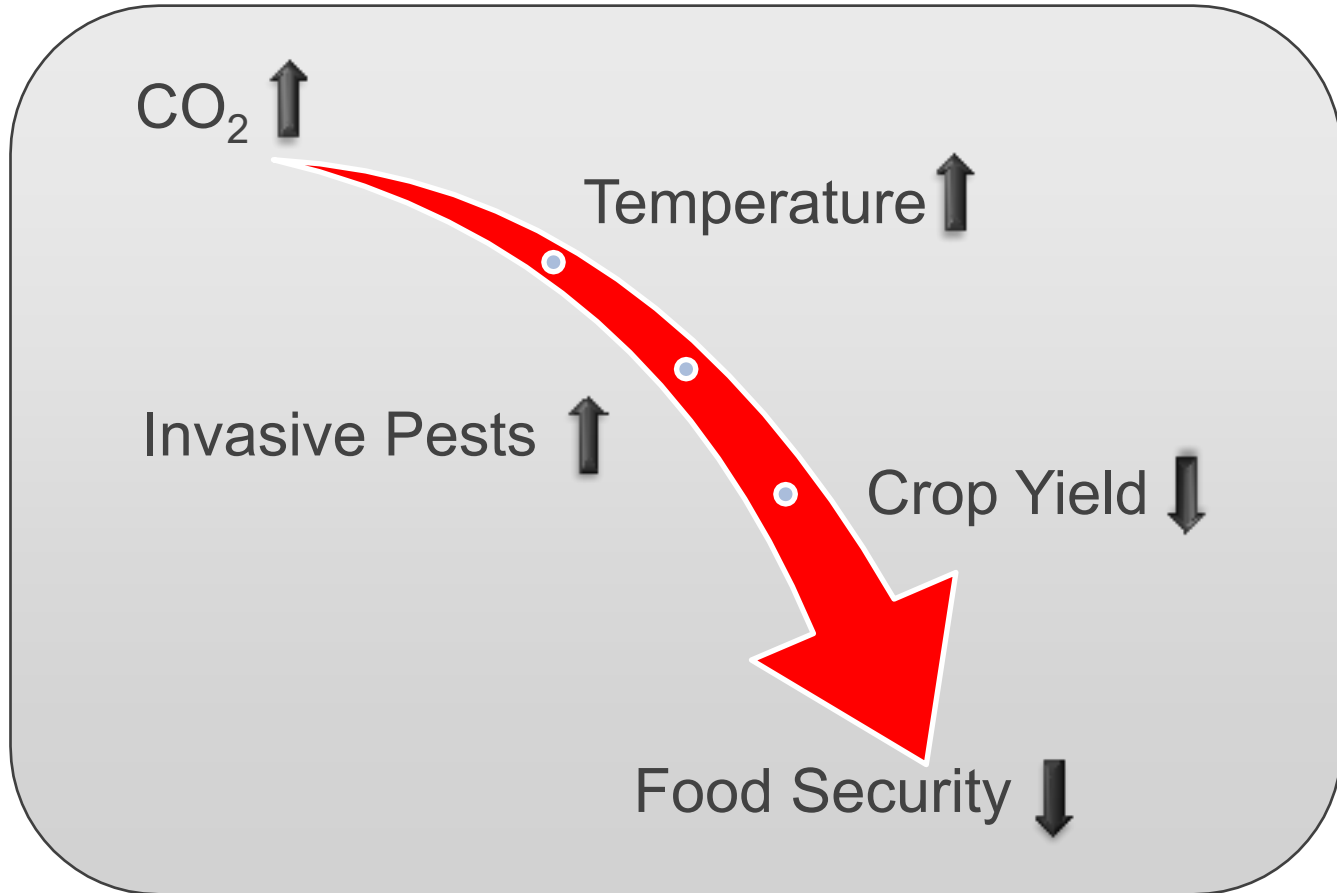
Encapsulation of Biologicals for Agriculture - Eco-Friendly Solutions for a Sustainable Future

Sustainable and Climate Resilient Food Production and Agriculture

March 29th, 2023

Team: *Brad Heater* (heater@battelle.org), Jonathon Clifton, Maria Fe Cruz Castillo, Kate Kucharzyk (kucharzyk@battelle.org), Veronica Fulwider, Anthony Duong, Mark Duffy, Jeff Cafmeyer, Sarah Ducceschi, and Colin Hinton

Climate Change Impacts Food Security



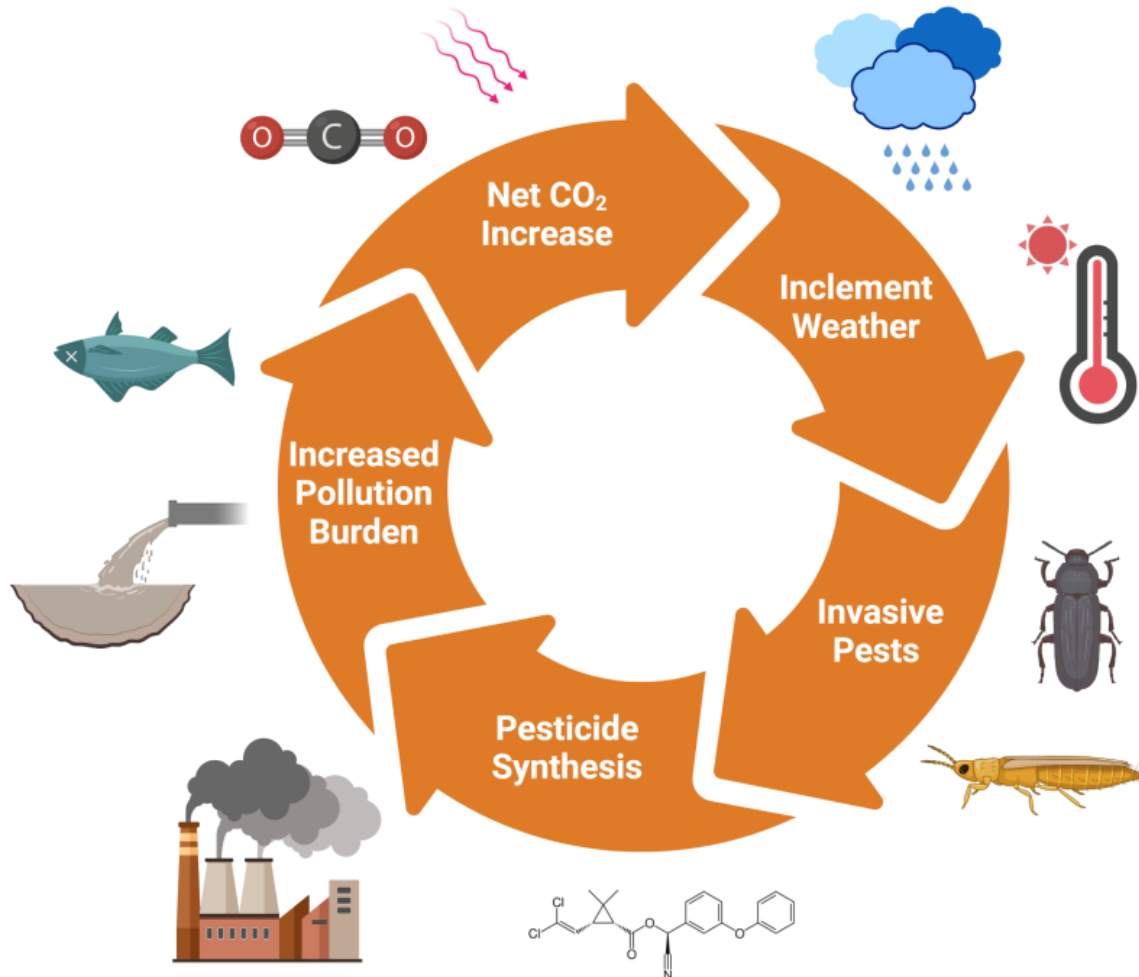
Credit: USDA/FAS/Curt Reynolds

The corn earworm



Credit: Anders Huseeth/NC State University

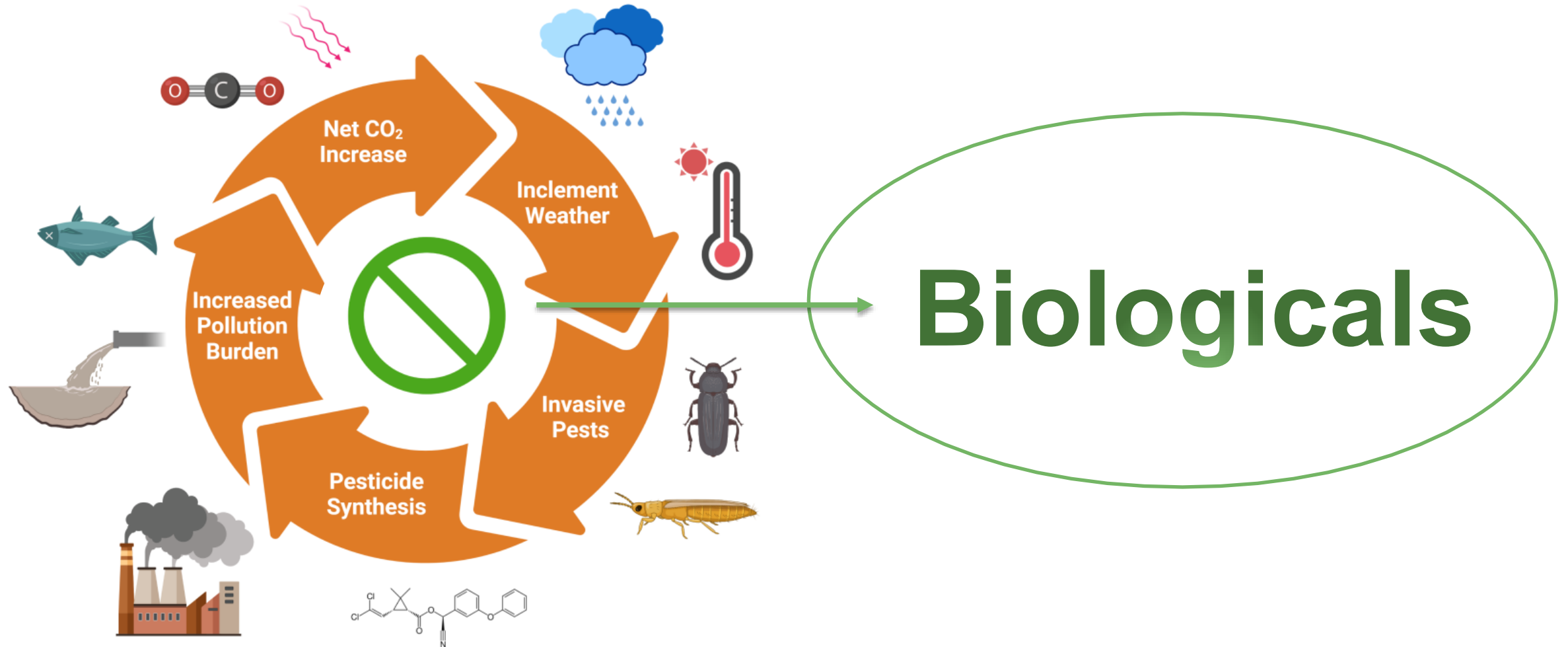
Perpetual Cycle of Climate Change in Agriculture



Created with BioRender.com

- Climate change results in rising temperatures which create stressful environments for crop fields
- Climate change alters the ecosystem, generating new niches for insects
- The extensive use of chemical pesticides to treat these new pests will increase the pollution burden as flooding intensifies
- The fossil fuels required to generate the billions of pounds of pesticide propagate the effects of climate change
- **So, what is the solution to stop this vicious cycle?**

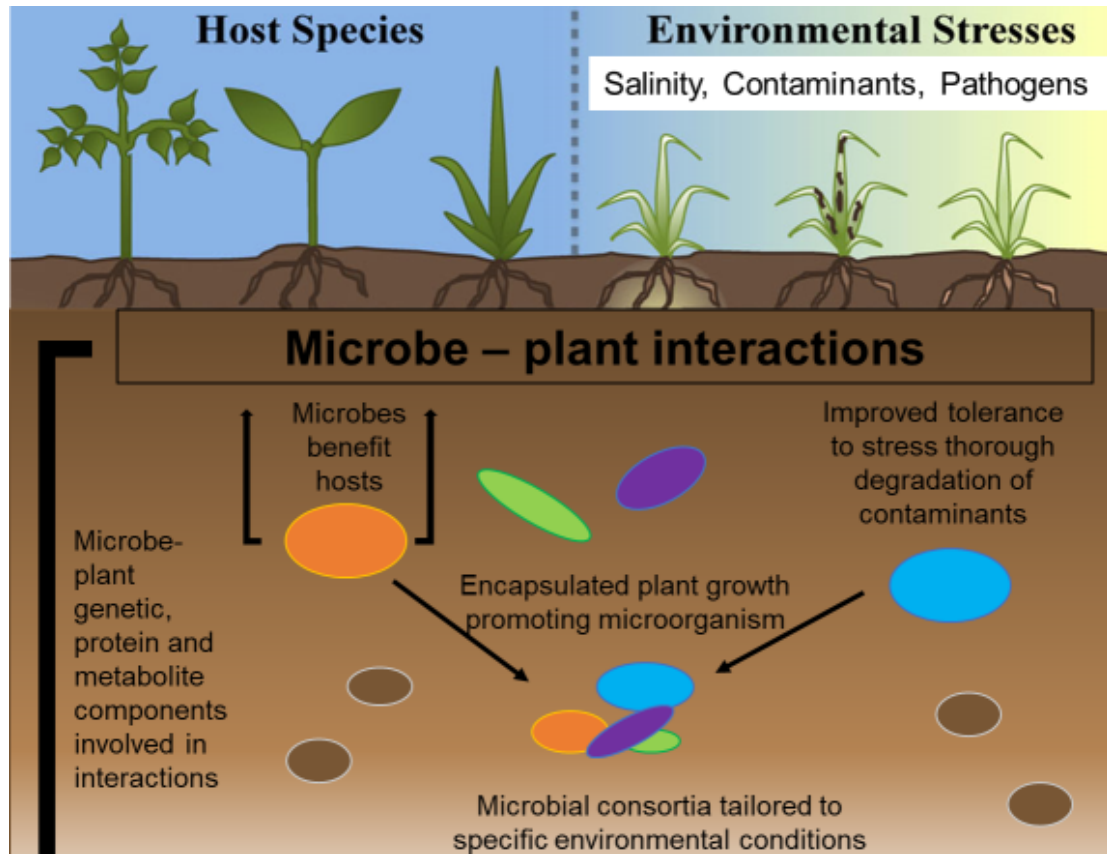
Perpetual Cycle of Climate Change in Agriculture



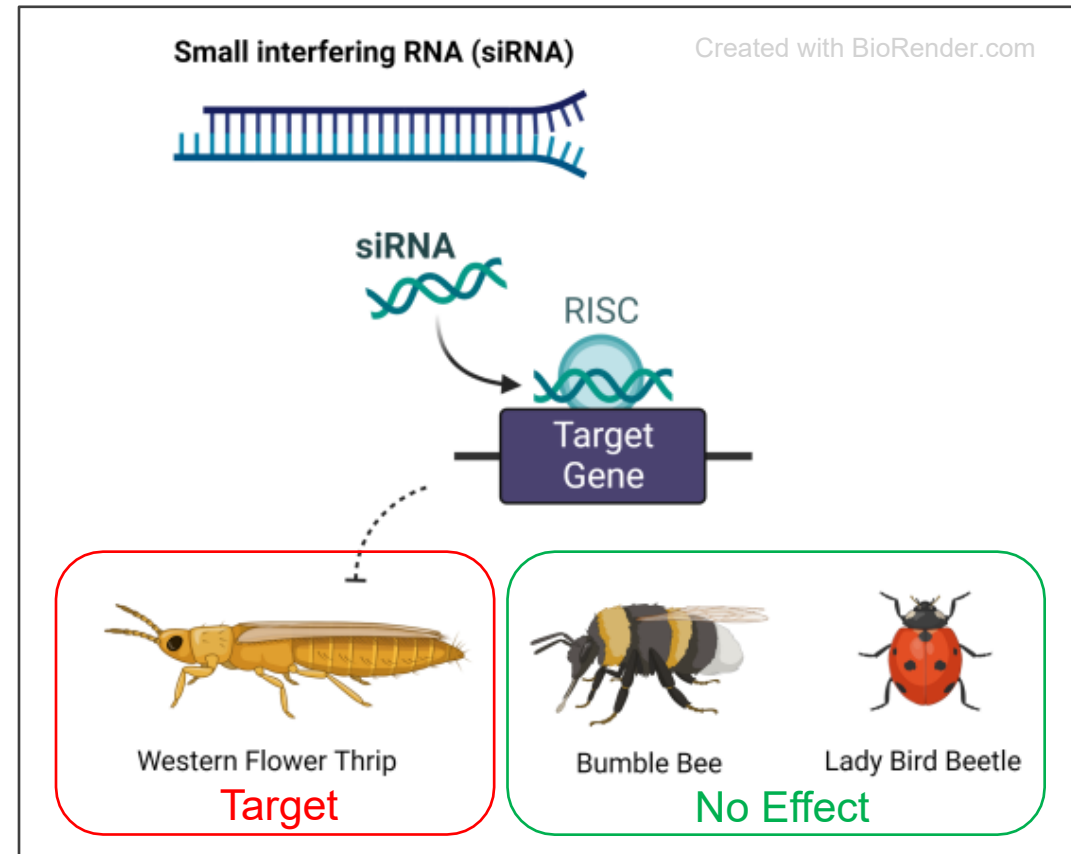
Created with BioRender.com

Biologicals in Agriculture

Microbes



Biomolecules

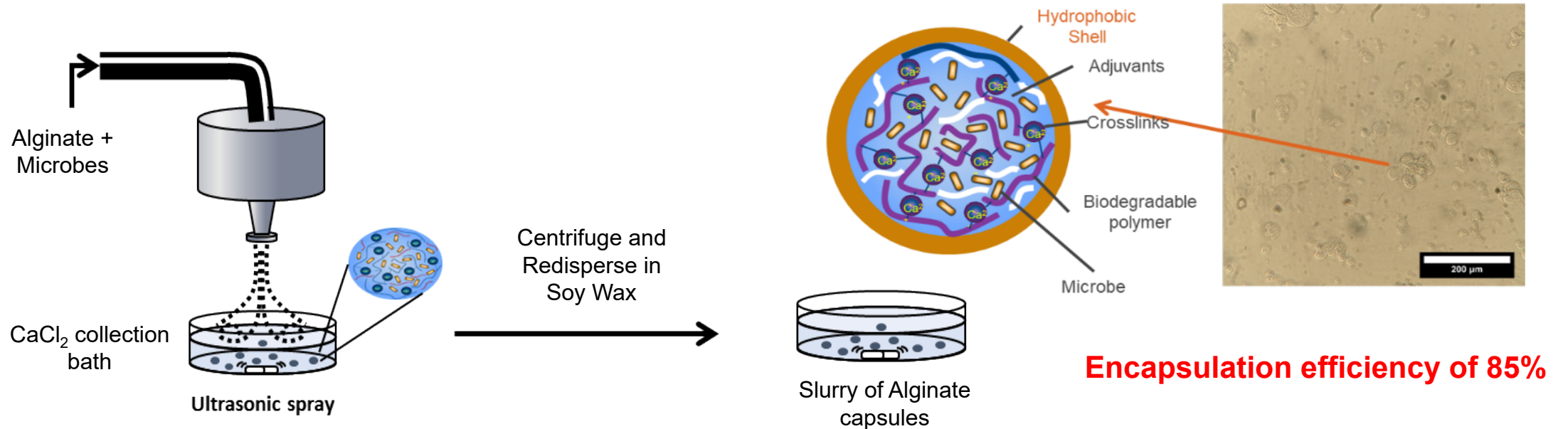


Challenge – Biologicals are significantly less stable than chemicals, so shelf-stability and field-longevity are real challenges in the development of biologicals in agriculture

Biostimulant Encapsulation

Superhydrophobic Water-Resistant Encapsulation (SWEL)
Technology

Generation of SWEL Capsules

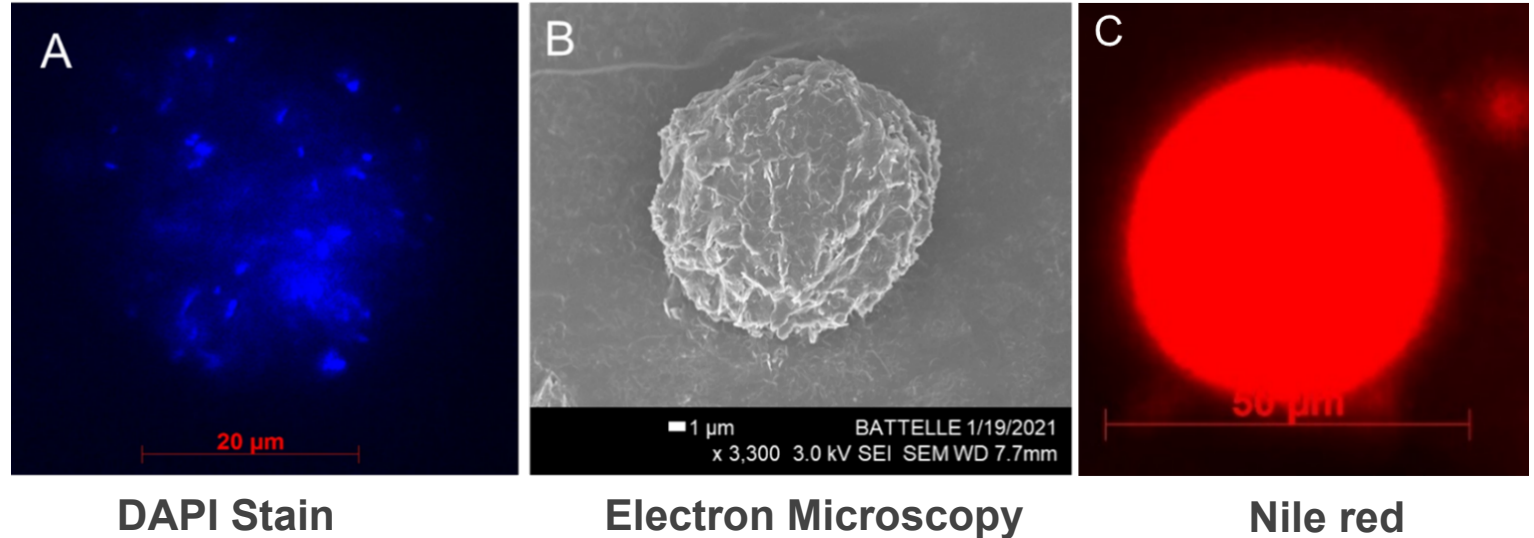


SWEL technology developed and compatible with calcium alginate encapsulation process to prevent back diffusion of Chemical Seed Coating Compounds (CSCs) and decrease desiccation of microbes during the shelf storage.

Kucharzyk *et al.* ACS Agricultural Science & Technology (2023). Under Revision.

SWEL Capsule Imaging

- *Pseudomonas protegens* was encapsulated in alginate and coated with soy wax to form SWEL
- Microbial cells were stained with DAPI to visualize DNA.
- Soy wax and lipids were stained with Nile Red

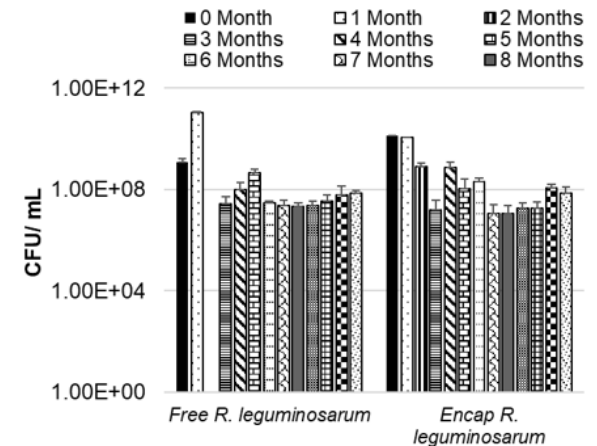
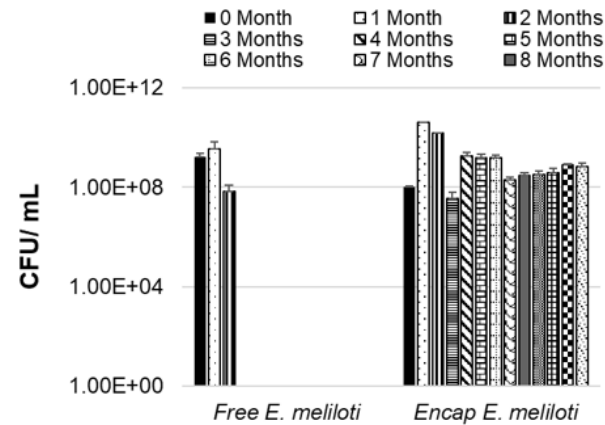
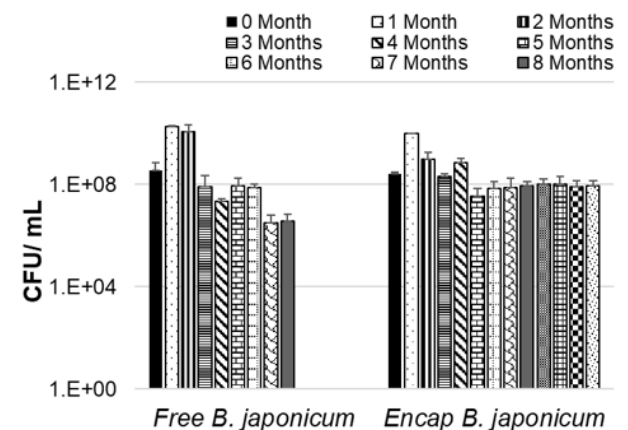
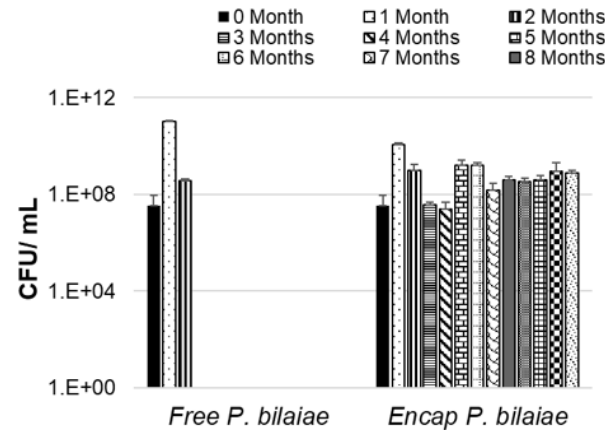


Capsules are round and show size distribution from 10-20 μm . This size is expected by agricultural formulators and aligns with expectations for product applied to the seed!

Kucharzyk *et al.* ACS Agricultural Science & Technology (2023). Under Revision.

Viability Testing – Storage Stability

- Encapsulation by SWEL can significantly improve storage stability
- *P. bilaiae* and *E. meliloti* demonstrate improved storage stability
- *B. japonicum* and *R. leguminosarum* show modest improvement in stability

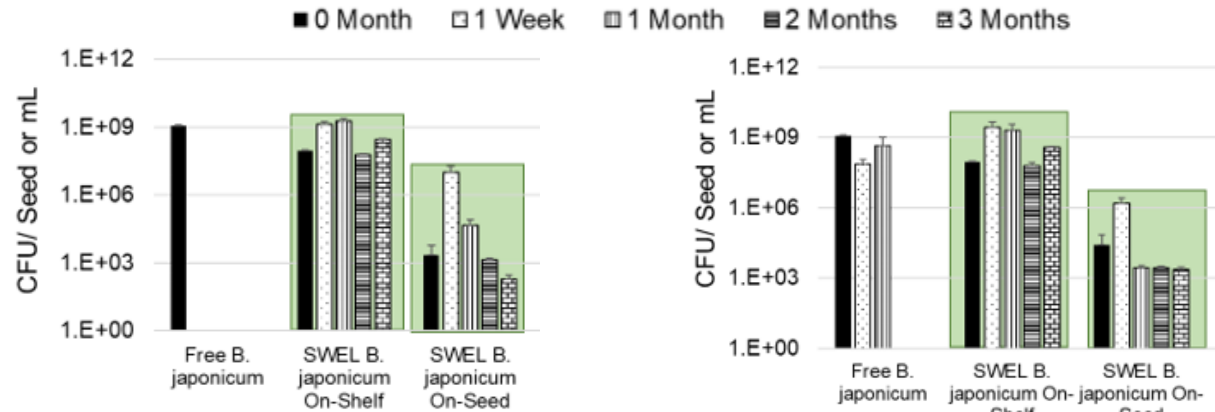


Kucharzyk et al. ACS Agricultural Science & Technology (2023). Under Revision.

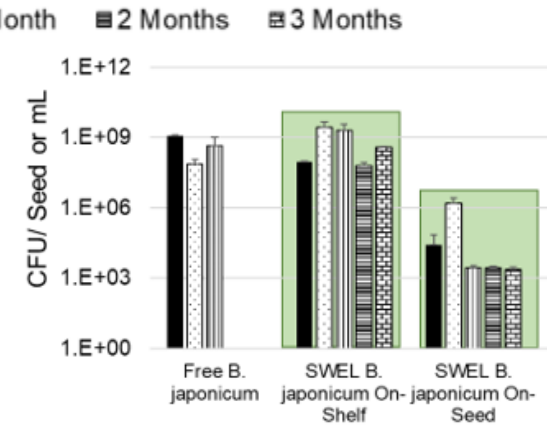
Viability Testing – CSC Compatibility

- Chemical Seed Coating Compounds (CSCs) help to:
 - Improve seed flow
 - Increase adhesion
 - Seed identification
 - Protection

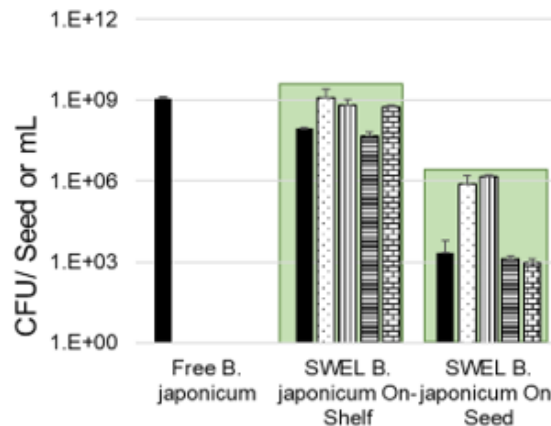
Genus/species	CSC Type	Test
B. japonicum strain (Buchanan) Jordan (ATCC, 10324)	Peridiam	On shelf and on-seed stability
	Flo Rite 1706	
	Pro-Ized Red Colorant	
	Poncho 600 FS	



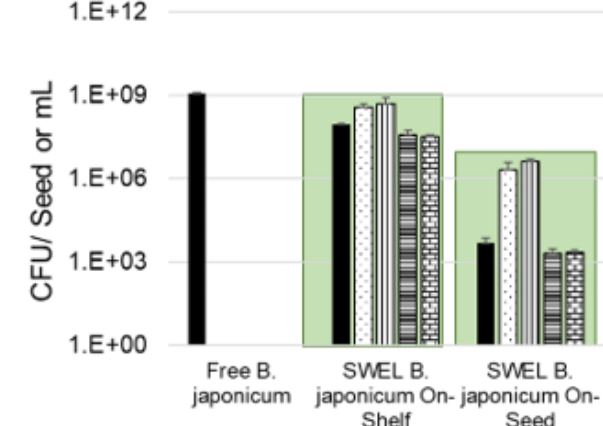
Peridiam



Flo Rite



Pro-Ized



Poncho 600

See Fadime Murdoch's poster #188 for greenhouse studies

Kucharzyk et al. ACS Agricultural Science & Technology (2023). Under Revision.

Biopesticide Encapsulation

Encapsulation of siRNA

Encapsulation of siRNA in a Protein Encapsulant

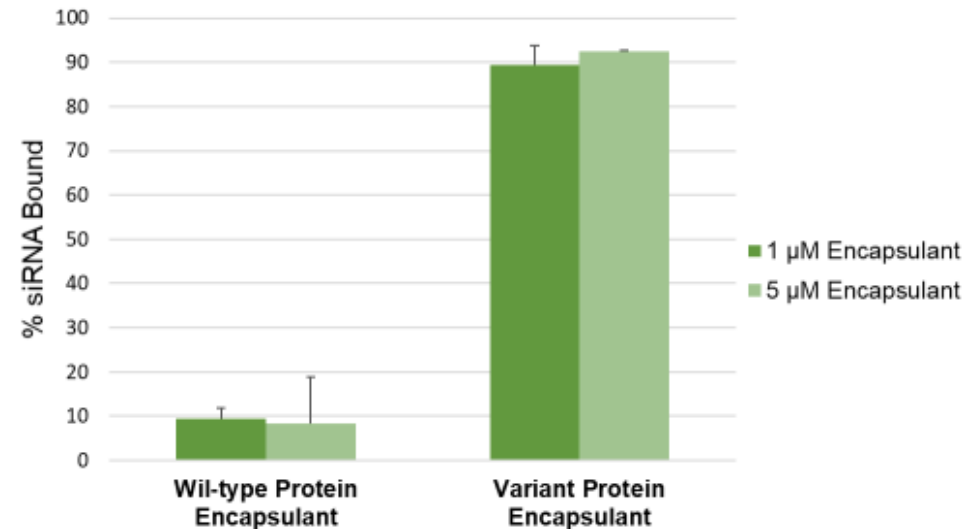
- **Key Objective – Generate a sprayable siRNA-based bioinsecticide for the agriculture industry**
 - Protein framework has been engineered to encapsulate siRNA with high efficiency
 - Delivery mechanism to insect midgut was established



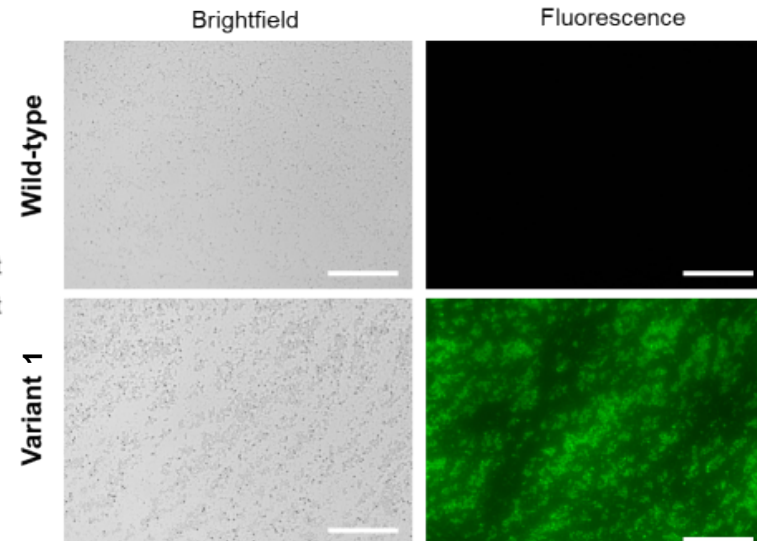
Western corn rootworm targeting siRNA labeled with fluorophore

Encapsulation

Variant 1

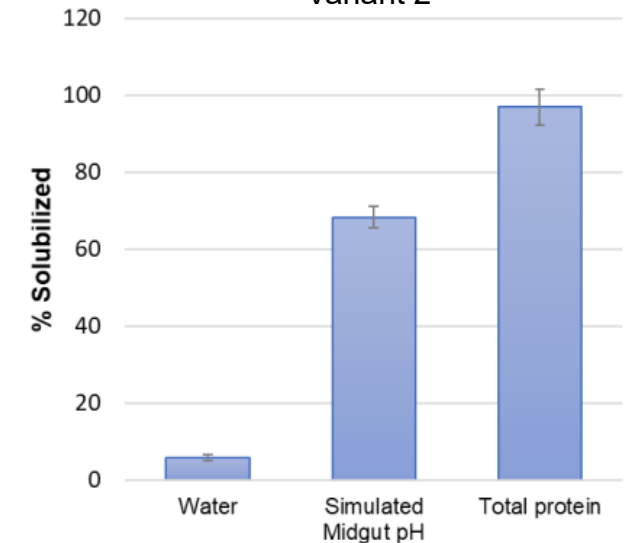


Microscopy



Release

Variant 2



Heater *et al.* Unpublished.

Project Technical Team

Encapsulation of siRNA Team



PI
Dr. Brad
Heater

Team Members

Megan Moore
Jonathon Clifton
Marian Fe Castillo
Sarah Dreher
Anurup Krishna

SWEL Team



PI
Dr. Kate
Kucharzyk

Team Members

Veronica Fulwider
Anthony Duong
Mark Duffy
Jeff Cafmeyer
Sarah Ducceschi
Colin Hinton

