Monica Alsup Dr. Spencer Arnesen\* Alex Durdello Patricia Gallardo Josh Johnson Ray Kannenberg\* Dr. Dan Lin Ketra Oketcho Anastasiia Onyshchenko\* Will Ranney\* Josh Sumner\* **Rhiannon Vargas** Marisa Yoder Alex Weil

Key former members: Dr. Mingsheng Qi\* Jeff Berry\* Dr. Kira Veley











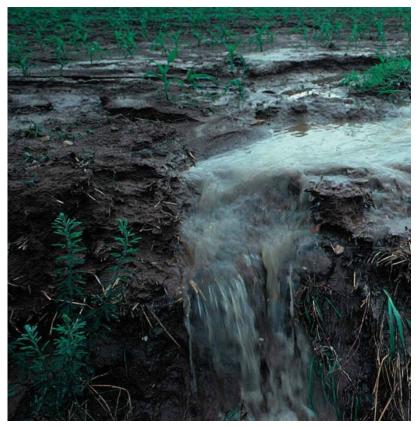
BILL& MELINDA GATES foundation

## Pollution from fertilizer runoff



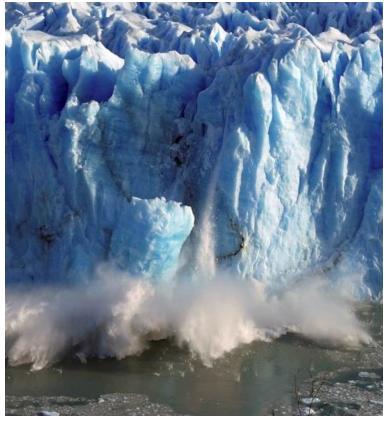
Source: NASA/Landsat/Phil Degginger/Alamy

### Soil degradation



Source: Lynn Betts/USDA NRCS

# Greenhouse gases and climate change



Source: Andres Forza / Reuters file

Modern day agriculture was not designed with sustainability in mind.

"Novel microbes and newly assembled syncoms to promote sustainable agriculture."







### Colonization and persistence

Major take homes:

- 1. Noise in data can be frustrating but can also help us learn about microbial systems.
- 2. We can't just skip to the end. Investment in foundational science is required to realize the potential benefit from microbial solutions in agriculture.

Despite significant potential, these products don't work as well as they could/need to.

WHY?







### Jeff Dangl (U. of North Carolina, Chapel Hill)

• SynComs developed based on associations with Arabidopsis

### Daniel Schachtman (U. of Nebraska, Lincoln)

• Sorghum grown in Nebraska under nitrogen or water limited conditions



### WHERE TO START?

• Can we translate findings from Arabidopsis into sorghum?

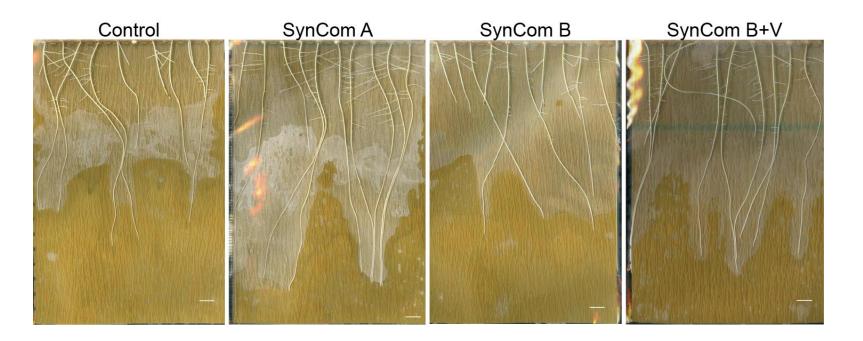
Article Published: 30 September 2020

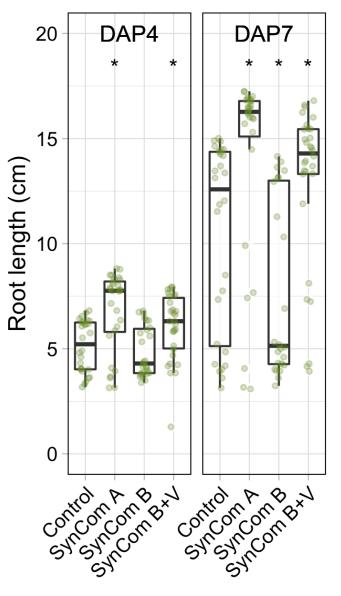
# A single bacterial genus maintains root growth in a complex microbiome

Omri M. Finkel, Isai Salas-González, Gabriel Castrillo, Jonathan M. Conway, Theresa F. Law, Paulo José Pereira Lima Teixeira, Ellie D. Wilson, Connor R. Fitzpatrick, Corbin D. Jones & Jeffery L. Dangl

- Specific strains of bacteria, eg. Arthrobacter, induce a short root phenotype in Arabidopsis.
- Variovorax strains can block the short root phenotype through the degradation of auxin.

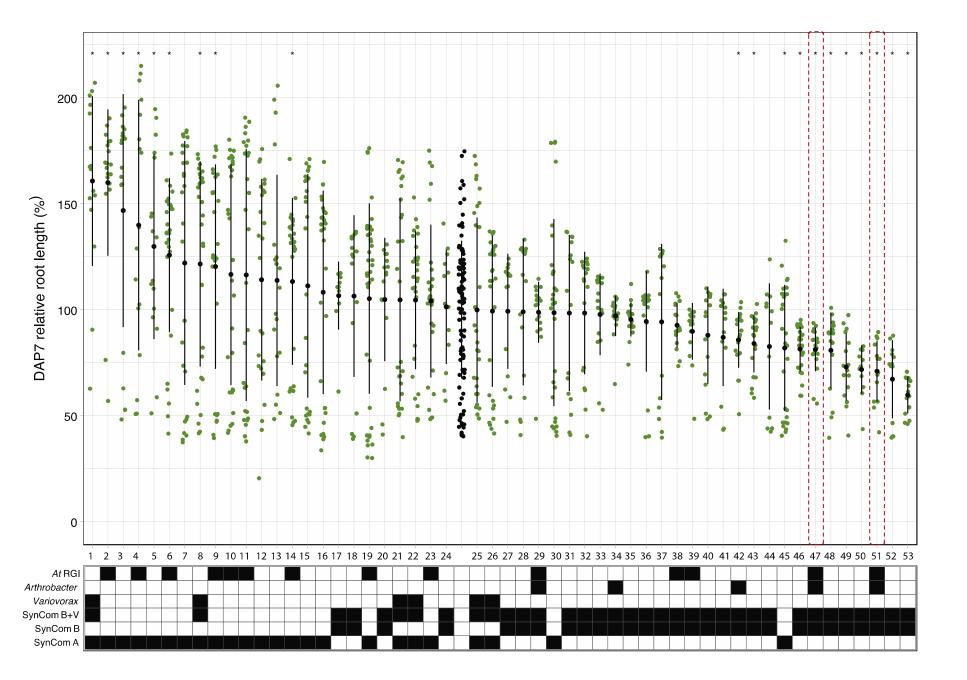
Sorghum seedling assay shows that strains of *Arthrobacter* induce a short root phenotype and that strains of *Variovorax* are protective.





- Effects between Arabidopsis and Sorghum are similar, but not exactly the same.
- 2. Even in a super controlled system, so much noise!





### WHY DO WE SEE SO MUCH NOISE?

Hypothesis: location of colonization might impact root phenotype.

Observation: Phenotype is dependent on location of colonization. Likely also dependent on the health of the seed.



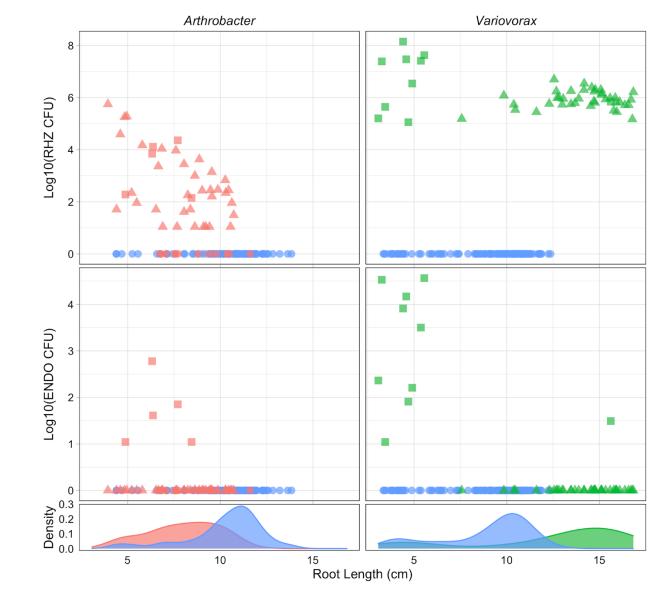
#### sample

- CL28 Arthrobacter
- CL14 Variovorax

Control

#### CFU

- RHZ\_CFU = 0, ENDO\_CFU = 0
- RHZ\_CFU > 0, ENDO\_CFU = 0
- RHZ\_CFU > 0, ENDO\_CFU > 0



Replication can help mitigate noise. Lemnatec phenotyper: 1,140 plants, weighed, watered and imaged every day.





Received: 14 September 2017 Accepted: 28 September 2017

DOI: 10.1002/pld3.23

#### **ORIGINAL RESEARCH**



## High-throughput profiling and analysis of plant responses over time to abiotic stress

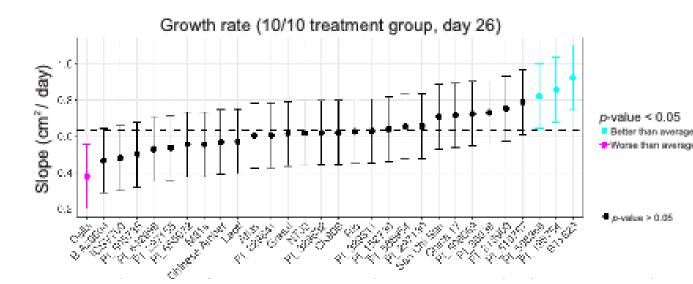
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Kira M. Veley<sup>1</sup> | Jeffrey C. Berry<sup>1</sup> | Sarah J. Fentress<sup>1</sup> | Daniel P. Schachtman<sup>2</sup> |
Ivan Baxter<sup>1,3</sup> | Rebecca Bart<sup>1</sup>
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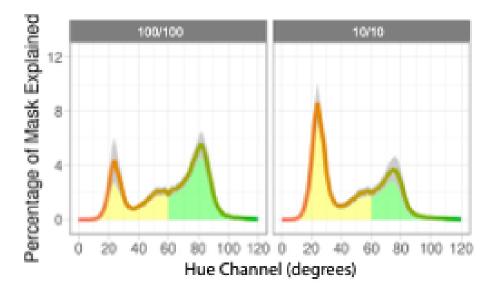
#### < BIOINFORMATICS AND GENOMICS

An automated, high-throughput method for standardizing image color profiles to improve imagebased plant phenotyping

Bioinformatics tool Bioinformatics Plant Science

Jeffrey C. Berry, Noah Fahlgren, Alexandria A. Pokorny, Rebecca S. Bart, Kira M. Veley





Do we see similar effects of SynComs in a more complicated system?

### PHENOTYPER METHODOLOGY:

Grow strains and resuspend in relatively equal parts in water.



Sterilize seeds and foam plugs and allow germination in presence of microbes

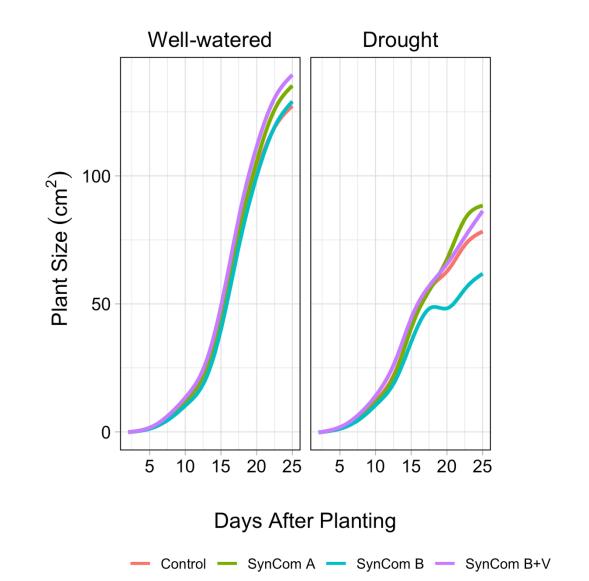


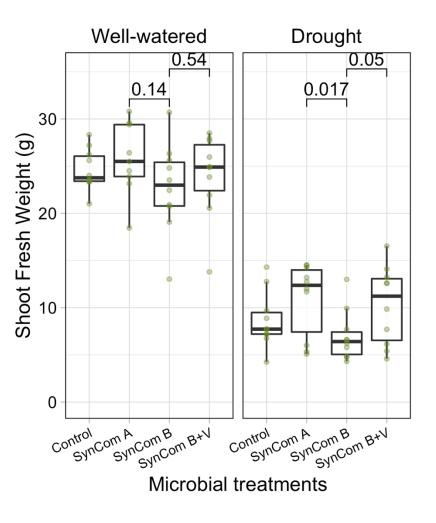


Transplant into tree pots after germination

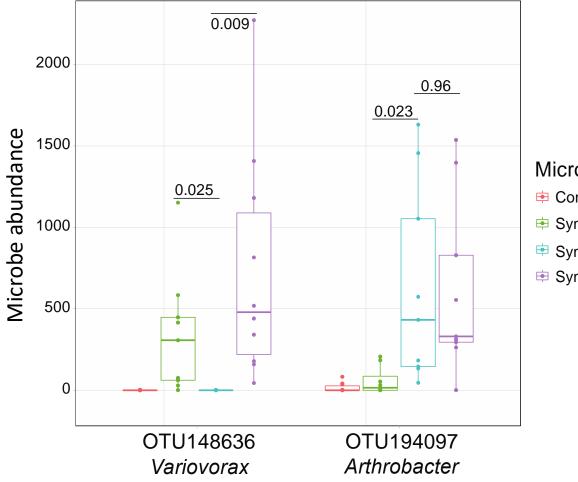


Variovorax strains can protect sorghum from drought stress, especially in the presence of Arthrobacter





### Colonization and persistence of Variovorax and Arthrobacter are inconsistent.



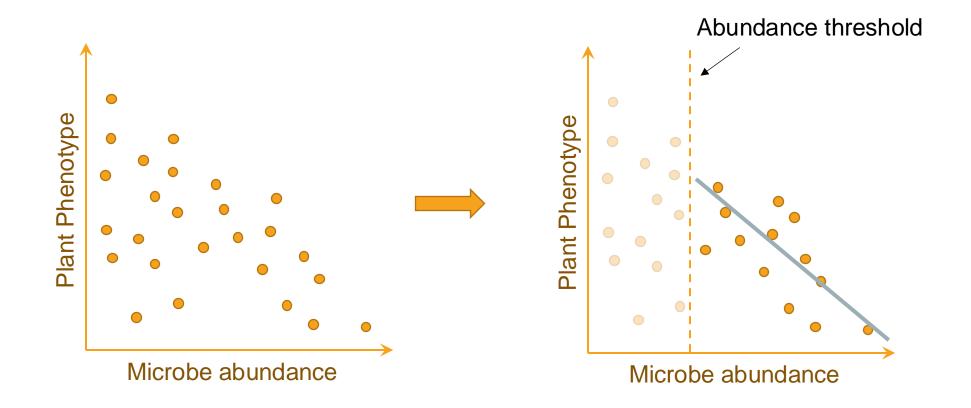




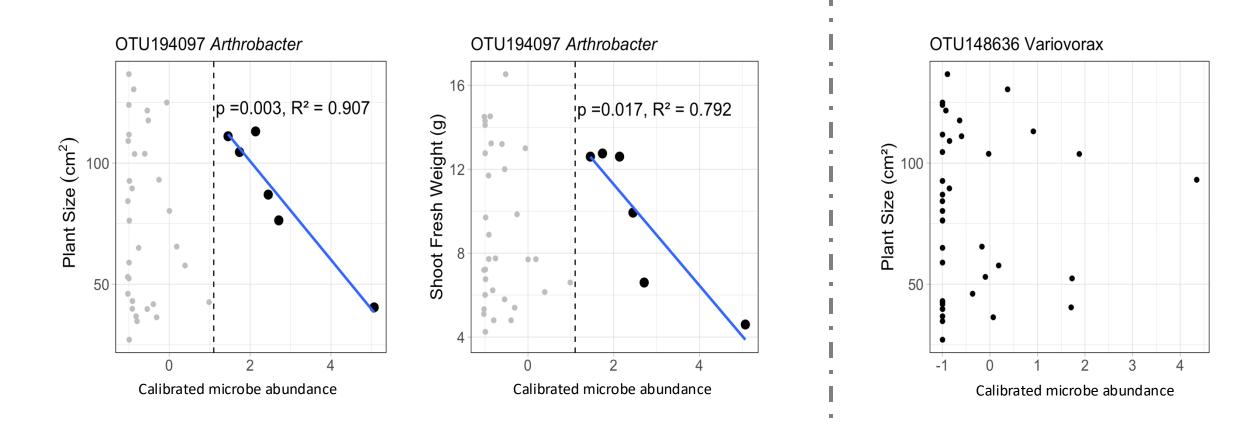
### Colonization and persistence

# Some microbes may only confer phenotype above a specific threshold abundance.

• Change-point models



For Arthrobacter, but not Variovorax, abundance correlates with plant phenotype.



н.

This was the beginning of the 'hinge model' analysis and 'biology informed logic models'





### Jeff Dangl (U. of North Carolina, Chapel Hill)

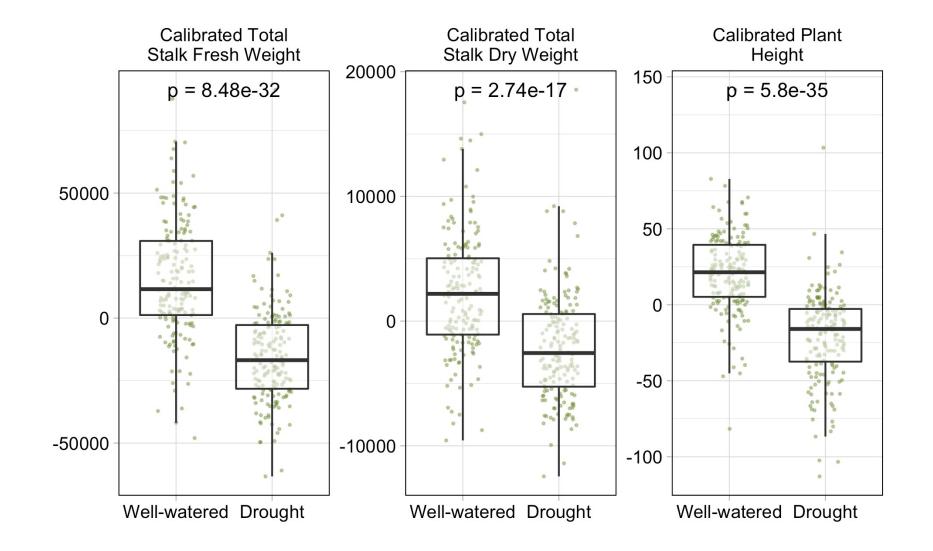
• SynComs developed based on associations with Arabidopsis

### Daniel Schachtman (U. of Nebraska, Lincoln)

• Sorghum grown in Nebraska under nitrogen or water limited conditions



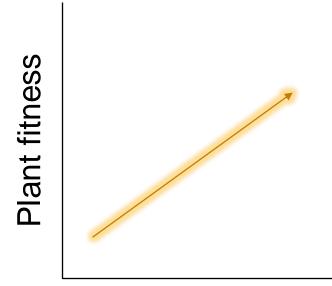
### Field data is very noisy!



Daniel Schachtman, UNL

Can we use the hinge model method to identify new symbiotic associations and the causal organisms from field experiments?

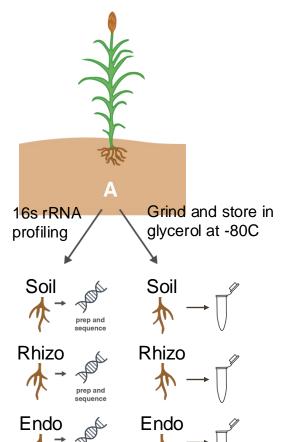
Foundational hypothesis: symbiosis between plant and microbe can be observed as a positive correlation between plant fitness and microbe abundance.



Microbe abundance

### Can we discover new symbioses between plants and bacteria?

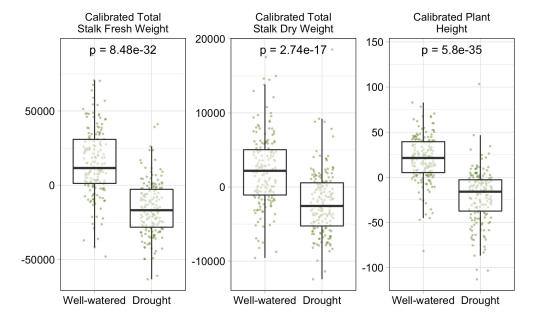
1. Sorghum plants were grown in the field under wellwatered or drought conditions.

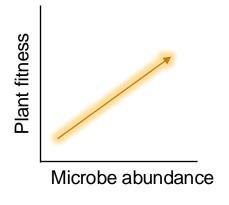


prep and

2. Diverse plant phenotypes quantified

3. Identify microbes whose abundance correlates with plant fitness.

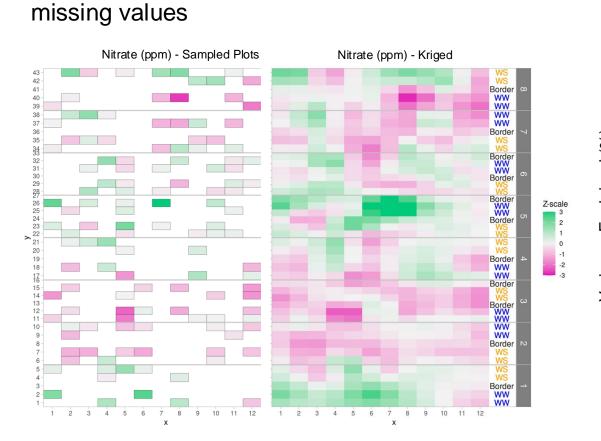




\*The first time we attempted this, we got no 'hits'

> Daniel Schachtman, UNL Susannah Tringe, JGI

## Method to account for (some) unintended experimental variation from field studies

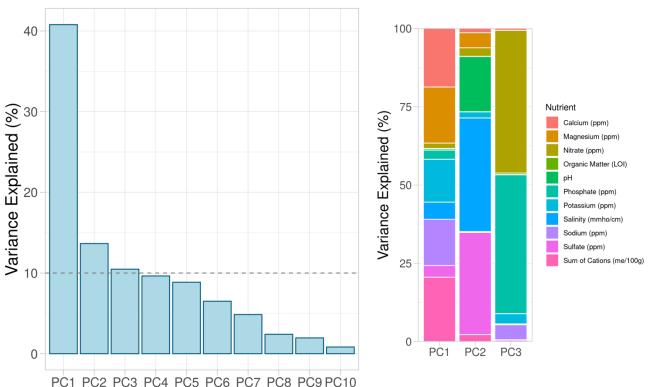


1) Measure soil properties & estimate

2) Dimension reduction through principal components

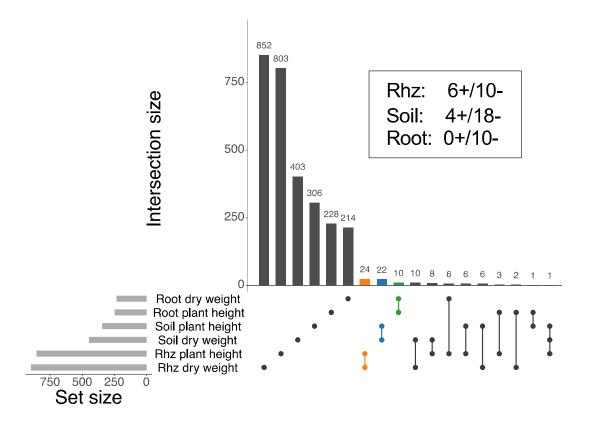


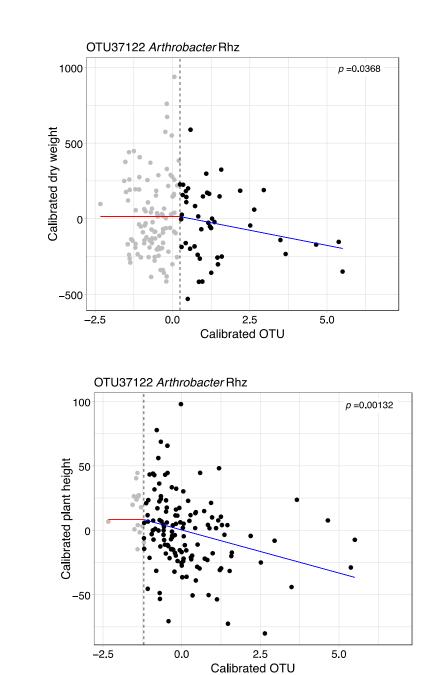
Jeff Berry Peng Liu (Iowa)



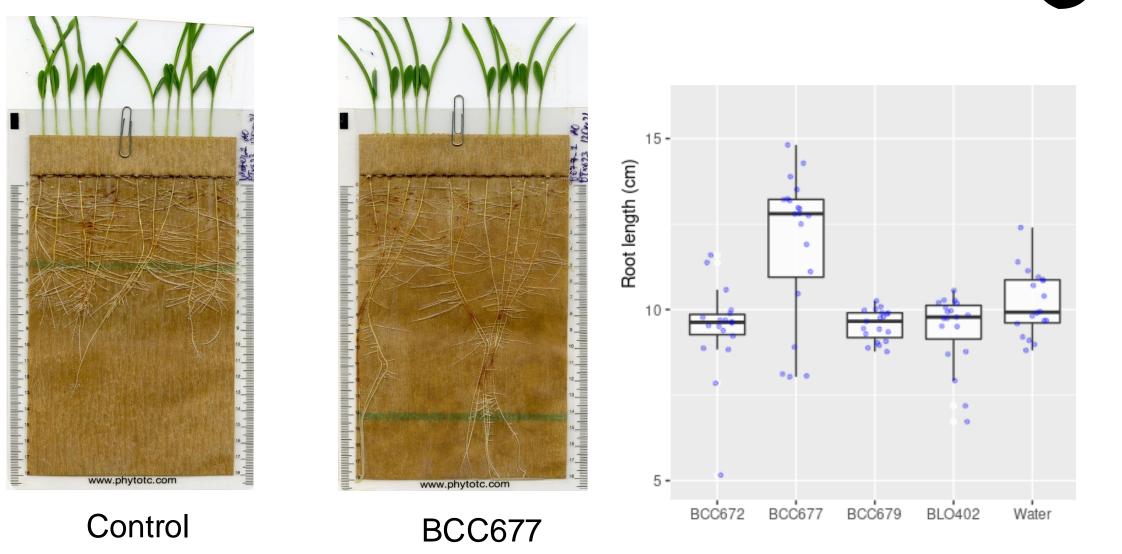
#### Berry et al 2022 eLife

Accounting for variation in soil properties revealed new candidates for microbes that affect plant phenotypes.

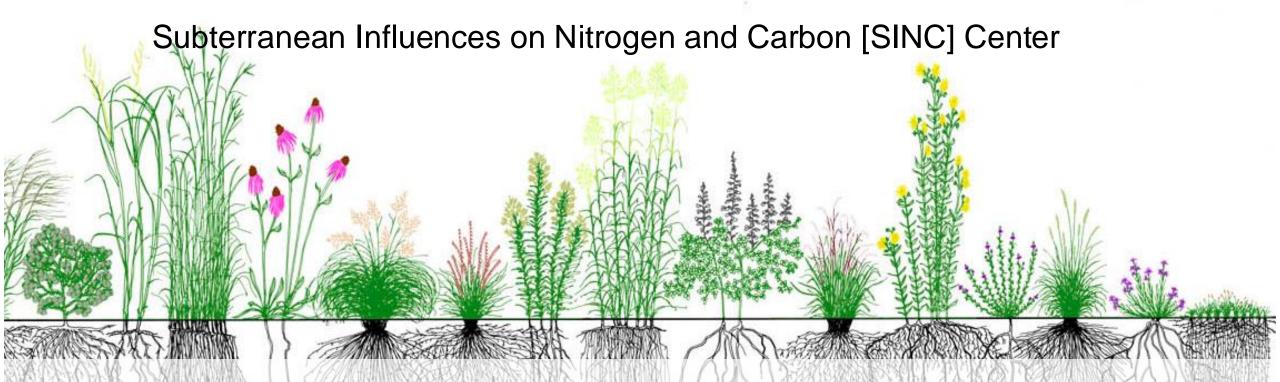




### Newly isolated strain results in increased root growth.



Anastasiia Onyshchenko

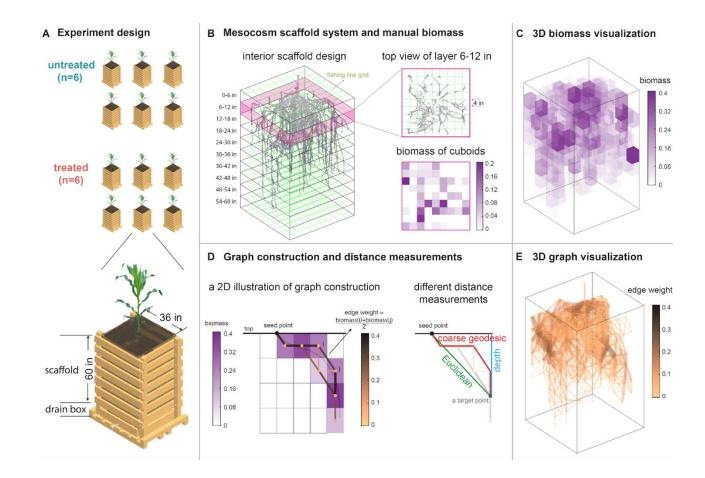


### **Overarching Center of Excellence concept:**

The stated goal of the SINC center is to decrease or eliminate the need for chemical fertilizer while maintaining high plant yields. This will be accomplished by promoting beneficial interaction with microbes, optimized plant genetics and sustainable cropping systems.

...with Doug Allen, Ivan Baxter, Armando Bravo and Chris Topp

## Larger controlled environment systems may reveal additional sources of noise in microbiome data.

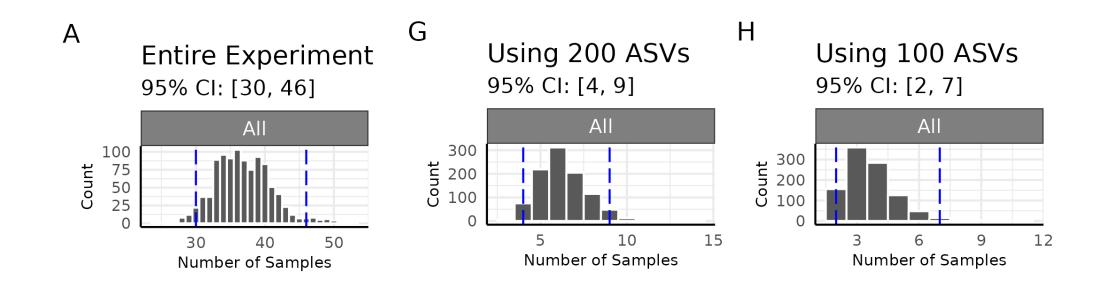




Mao Li, Chris Topp and lab.

Is the root microbiome consistent across the root architectural system?

If not, how many root samples do you need to sample, to capture the microbial diversity across the root architectural system?



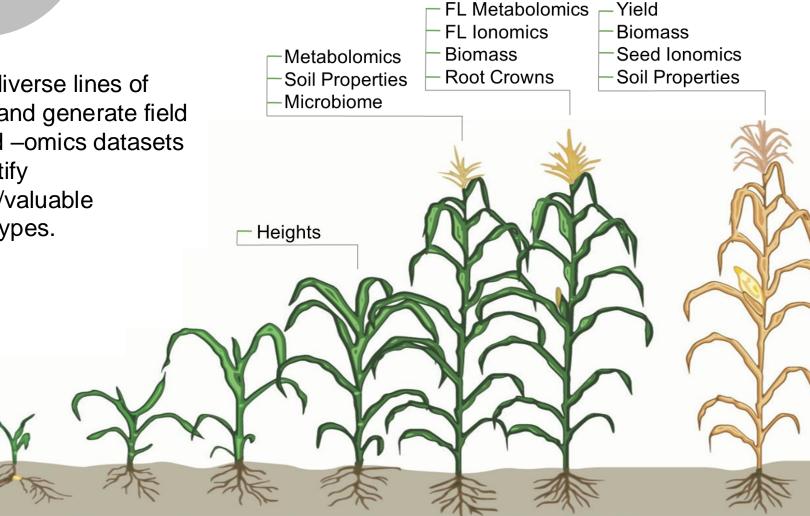
Diverse microbiomes exist across a mature root architecture and this likely contributes to additional noise.

Under review Mao Li and Josh Sumner



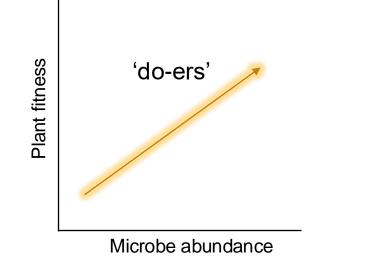
### NOVEL PLANT TRAITS THAT GOVERN NITROGEN USE EFFICIENCY AND/OR CULTIVATING A HEALTHY MICROBIOME.

Grow diverse lines of maize and generate field derived –omics datasets to identify unique/valuable phenotypes.

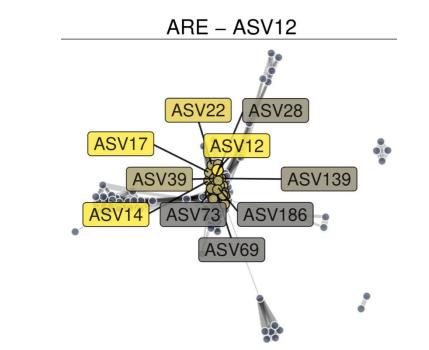




### **Biology Informed Logic Models**



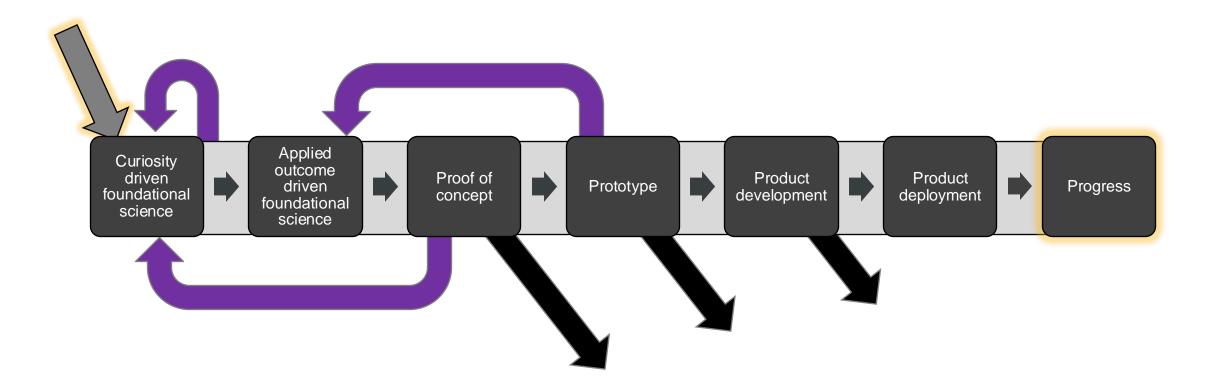
Symbiosis between plant and microbe can be observed as a positive correlation between plant fitness and microbe abundance.



Co-occurrence can be used as an indicator of candidate 'helpers'. These patterns may help in assembly of SynComs.

Spencer Arnesen

### PIPELINE TO PROGRESS



Foundational work fuels the pipeline. Projects sometimes exit the pipeline for good reasons. "Novel microbes and newly assembled syncoms to promote sustainable agriculture."







### Colonization and persistence

Major take homes:

- 1. Noise in data can be frustrating but can also help us learn about microbial systems.
- 2. We can't just skip to the end. Investment in foundational science is required to realize the potential benefit from microbial solutions in agriculture.

## Questions?



beckybartlab.org