

Development of Seed-Treating Biostilumants for Promoting Soybean Growth and Health

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Challenges in Crop Production



https://www.pioneer.com/us/agronomy/soybean_fertility.html



- Low soil fertility
- Biotic and abiotic stresses
- High cost of fertilizers and pesticides
- Resistance to pesticides



<https://extension.sdstate.edu/late-season-soybean-diseases-know-whats-killing-your-soybeans>

→ *It is imperative to develop innovative materials and methods for more sustainable production of crops*

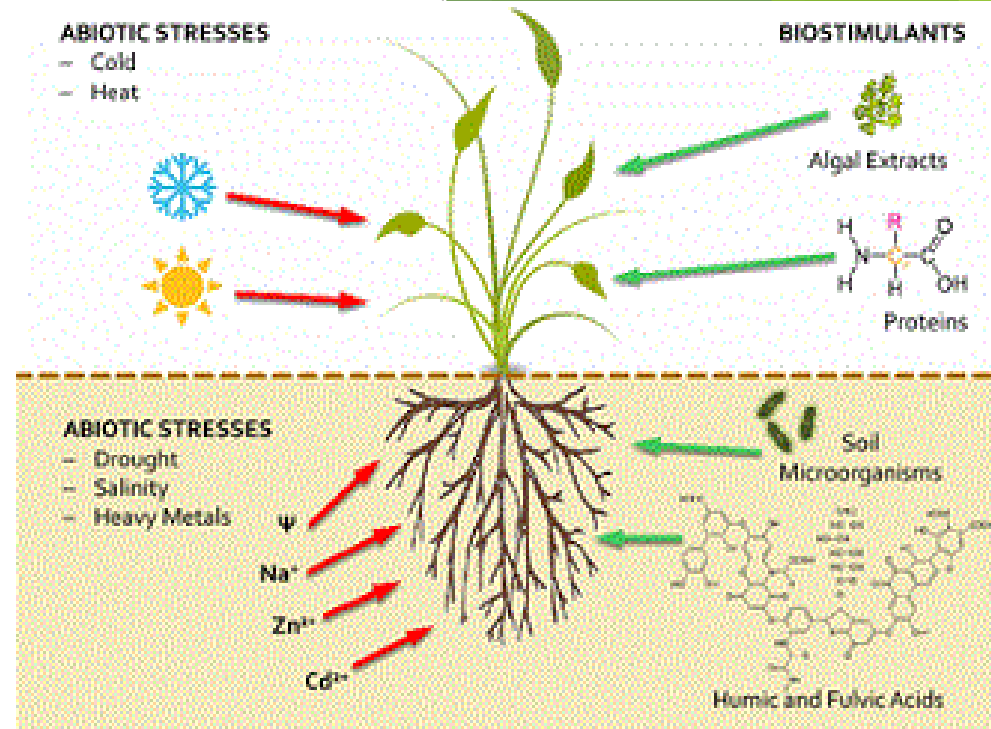
Two Major Directions of This Study

I. Biostimulants

- (Definition) Any natural substances, microorganisms, or mixtures of them that enhance nutrition efficiency, growth, yield, quality, and/or tolerance to biotic and abiotic stresses.
- Contribute to sustainable and high-output/low-input crop productions

II. Seed Treatment

- More efficient and economical compared to other application methods, such as foliar spraying and soil drenching.

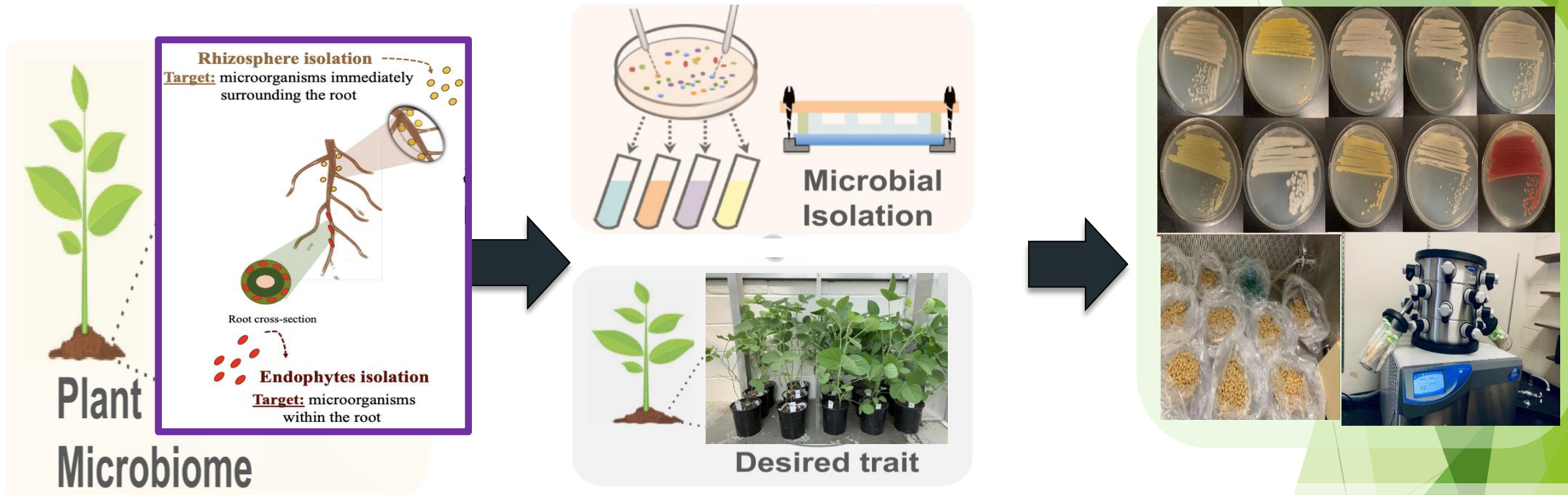


(Image from 'Bio Based Press')



(Image from 'CropLife')

Conceptual Workflow



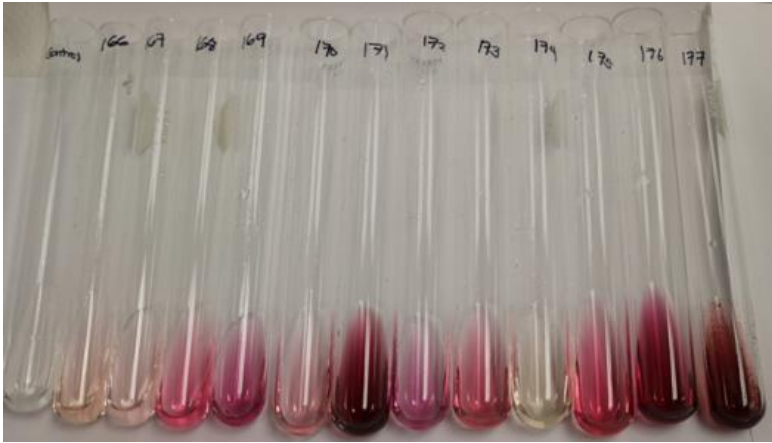
- Isolation of 'soybean-associated beneficial bacteria (SABB) from conspicuously healthy or disease tolerant plants in soybean fields

- Screening based on various beneficial activities
- Compatibility test and formulation of SABB consortia
- Test efficacy on soybean plants

- Characterization of biological mechanisms
- Development of formulation methods for seed or soil treatments

Beneficial Activities for Screening SABB

IAA production



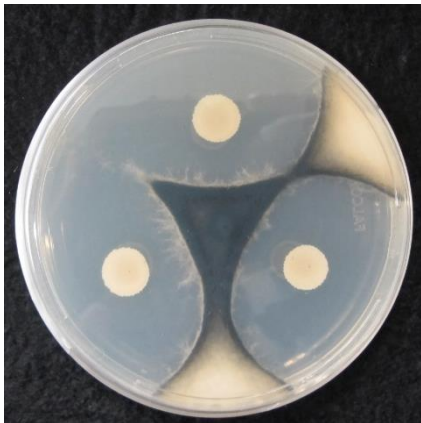
Germination and seedling growth



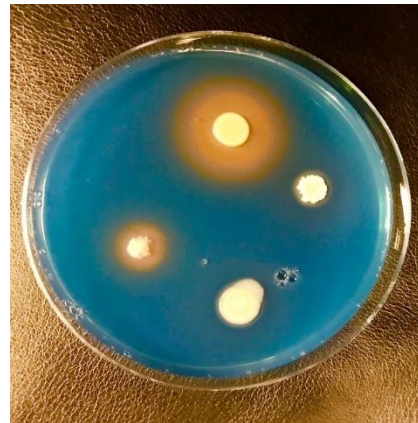
Nitrogen fixation



Antimicrobial activities



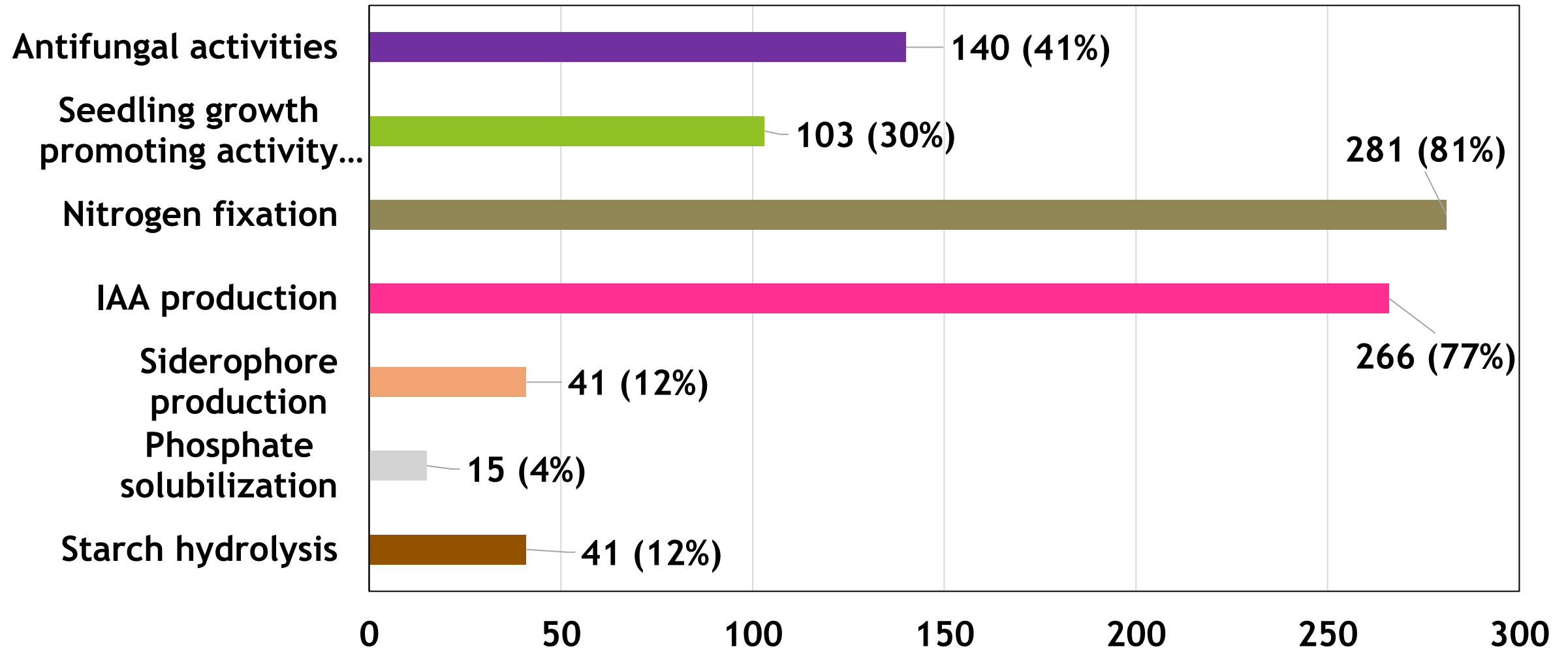
Siderophore production



Phosphate solubilization



Beneficial Activities of the Screened SABB



SABB Consortia and Their Subsets Tested

SABBset1 (9 members)
SABBset2 (14 members)
SABBset3 (23 members)

SABBmin1 (5 members)
SABBmin2 (7 members)
SABBmin3 (9 members)
SABBmin4 (10 members)

SABB consortia	Composition (members)	Antifungal activities	Growth promoting activity	Nitrogen fixation	IAA production	Siderophore production	Phosphate solubilization
Set1	9						
Set2	14						
Set3	23						
Setmin1	5						
Setmin2	7						
Setmin3	9						
Setmin4	10						

Greenhouse tests for treated soybean seeds (biomass)



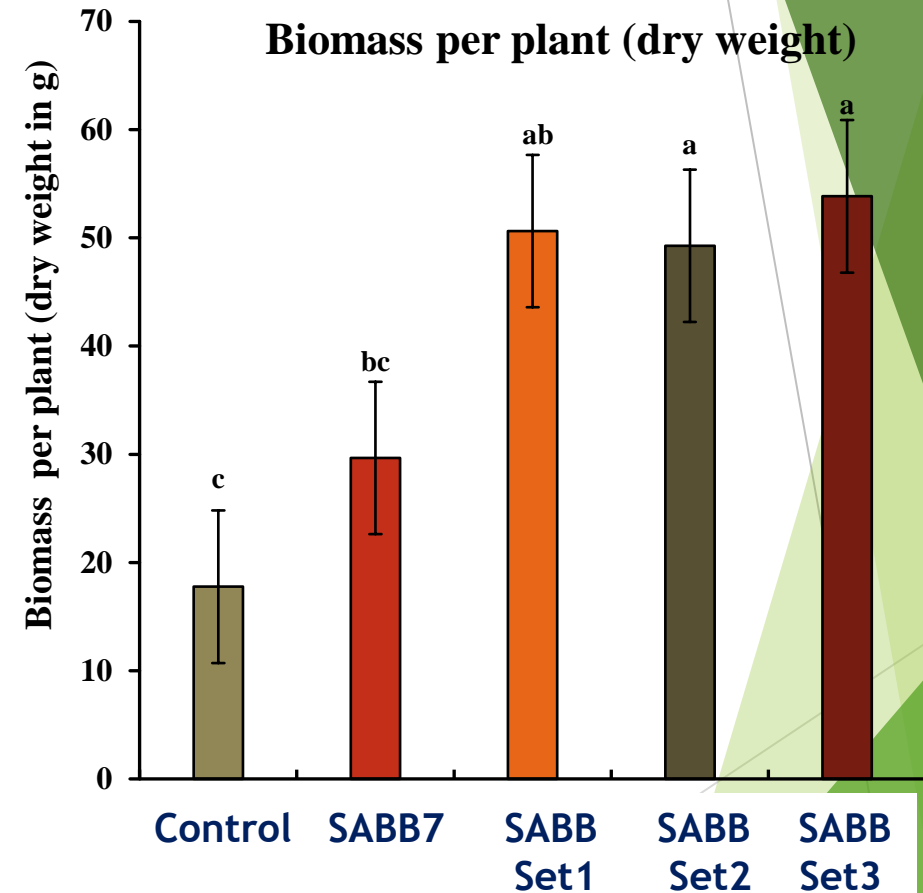
Control

SABB7

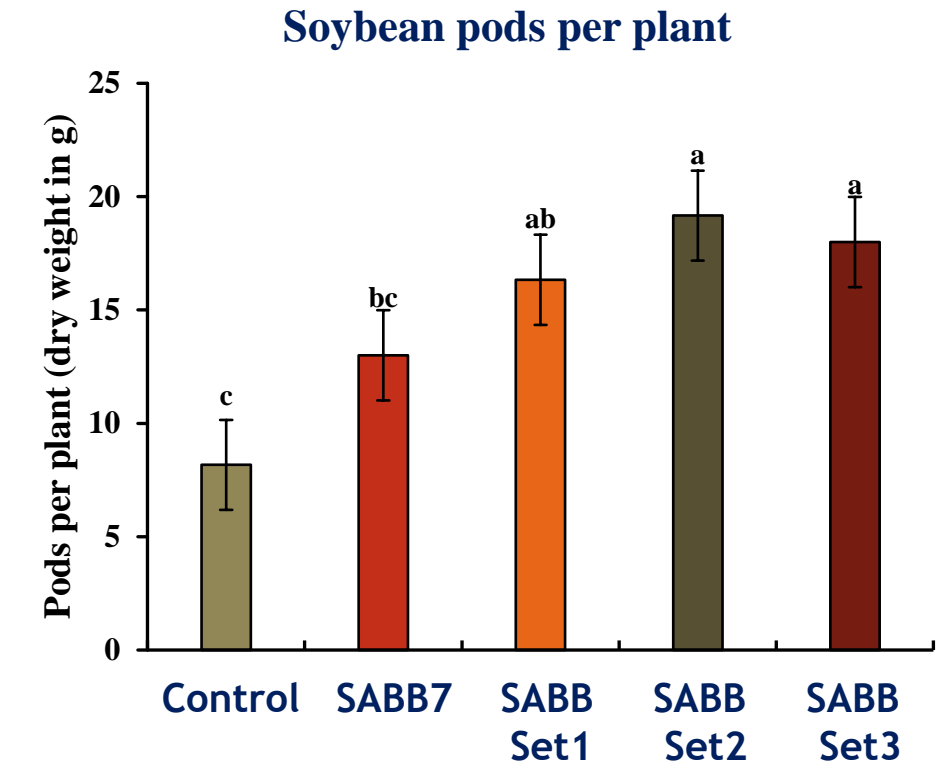
SABB
Set1

SABB
Set2

SABB
Set3




Greenhouse tests for treated soybean seeds (pods)



SABB Consortia and Their Subsets Tested

SABBset1 (9 members)
SABBset2 (14 members)
SABBset3 (23 members)

SABBmin1 (5 members)
SABBmin2 (7 members)
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Set1	 9						
Set2	14						
Set3	23						
Setmin1	5						
Setmin2	7						
Setmin3	9						
Setmin4	10						

Seedling growth resulted from subsets of SABBs



Untreated
Control

Apron
Maxx

SABB
Set1



Untreated
Control

Apron
Maxx

SABB
Set2



Untreated
Control

Apron
Maxx

SABB
Set3



Untreated
Control

Apron
Maxx

SABB
Min1



Untreated
Control

Apron
Maxx

SABB
Min2

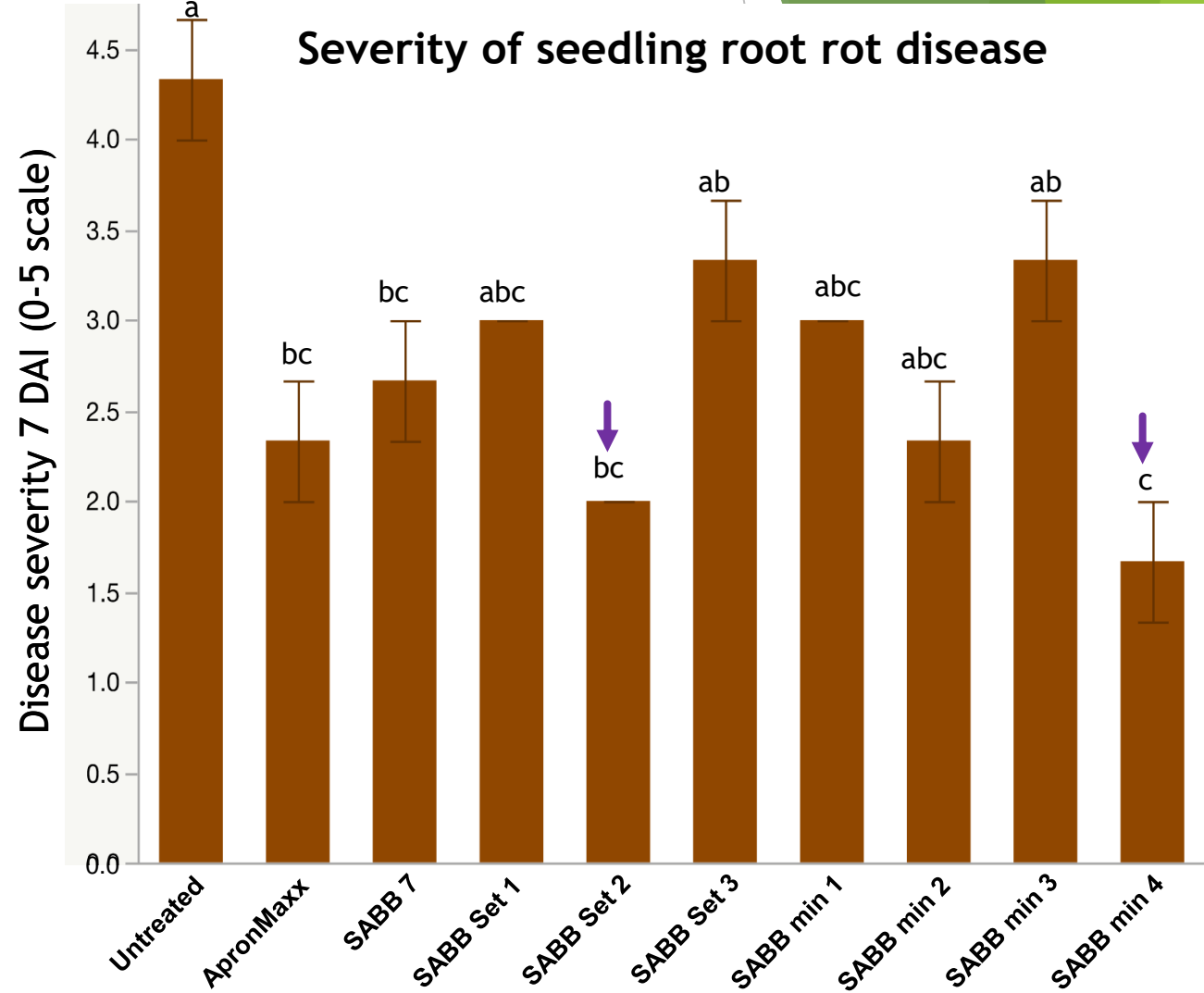


Untreated
Control

Apron
Maxx

SABB
Min4


Effect of SABBs on Seedling root rot (*Rhizoctonia solani*)



SABB Consortia and Their Subsets Tested

→ SABBset1 (9 members)
SABBset2 (14 members)
SABBset3 (23 members)

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Field tests conducted at three research stations of LSU AgCenter



**Doyle Chambers Central
Research Station, Baton Rouge
(July 1, 2021)**

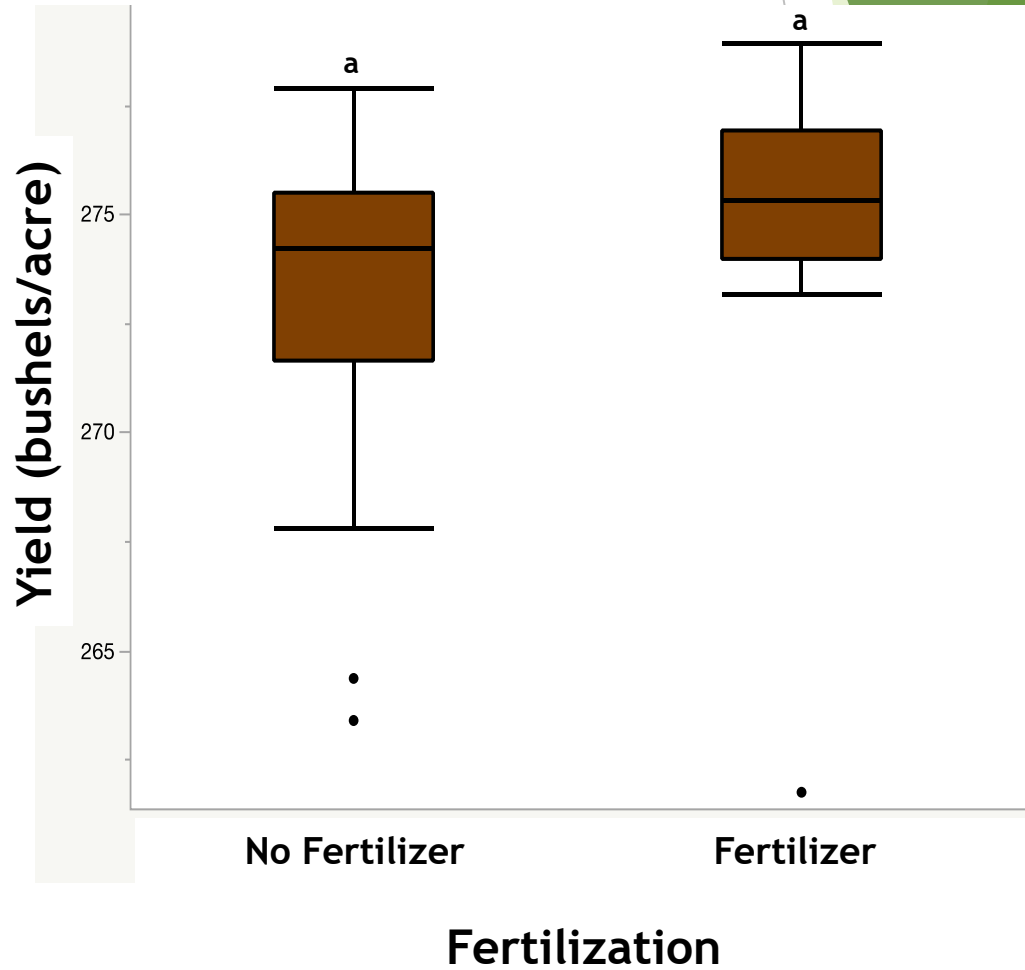
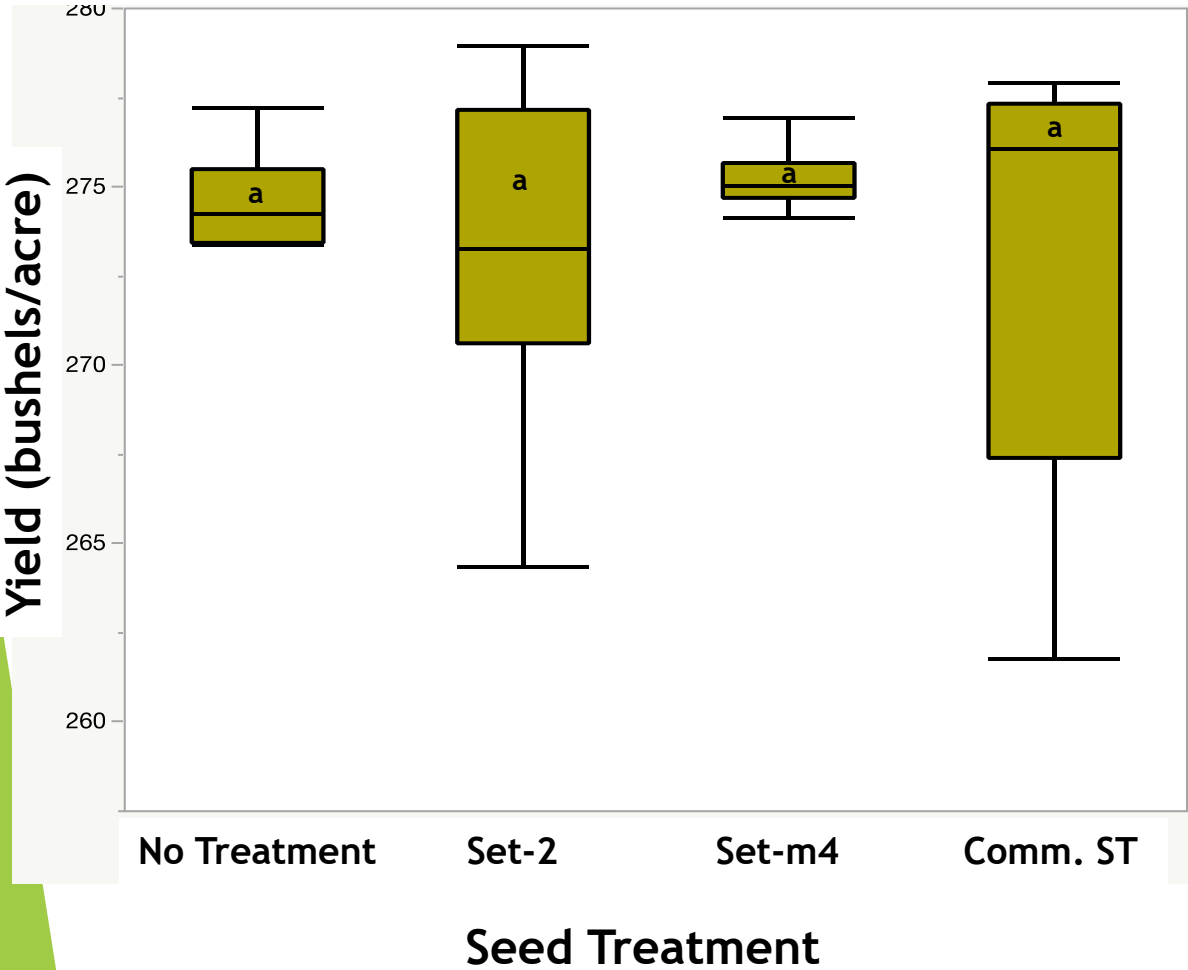


**Dean Lee Research Station,
Alexandria (July 21, 2021)**

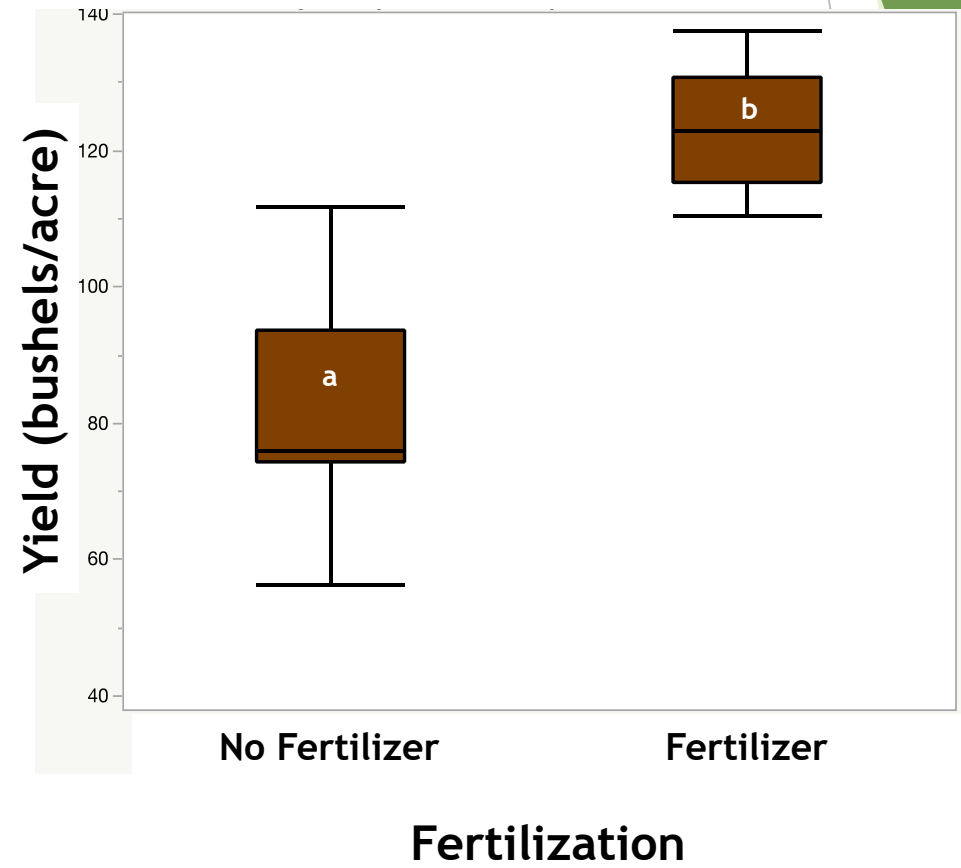
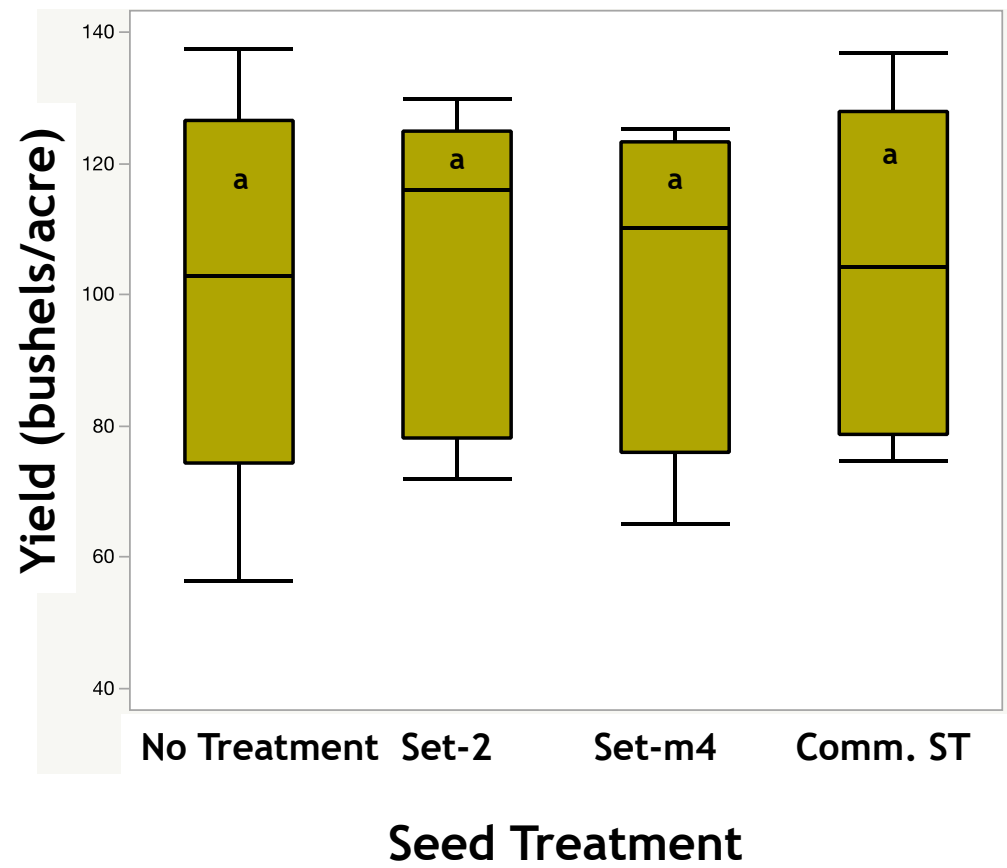


**Red River Research Station,
Bossier City (August 12,
2021)**

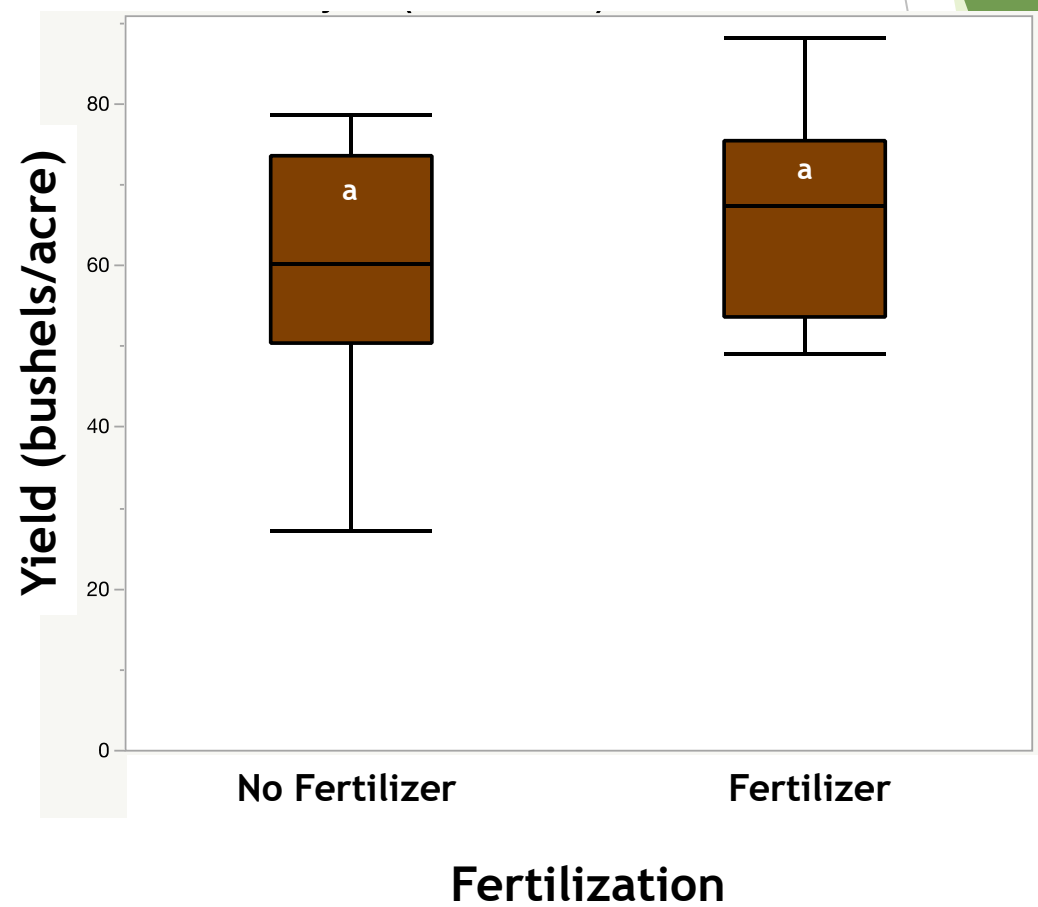
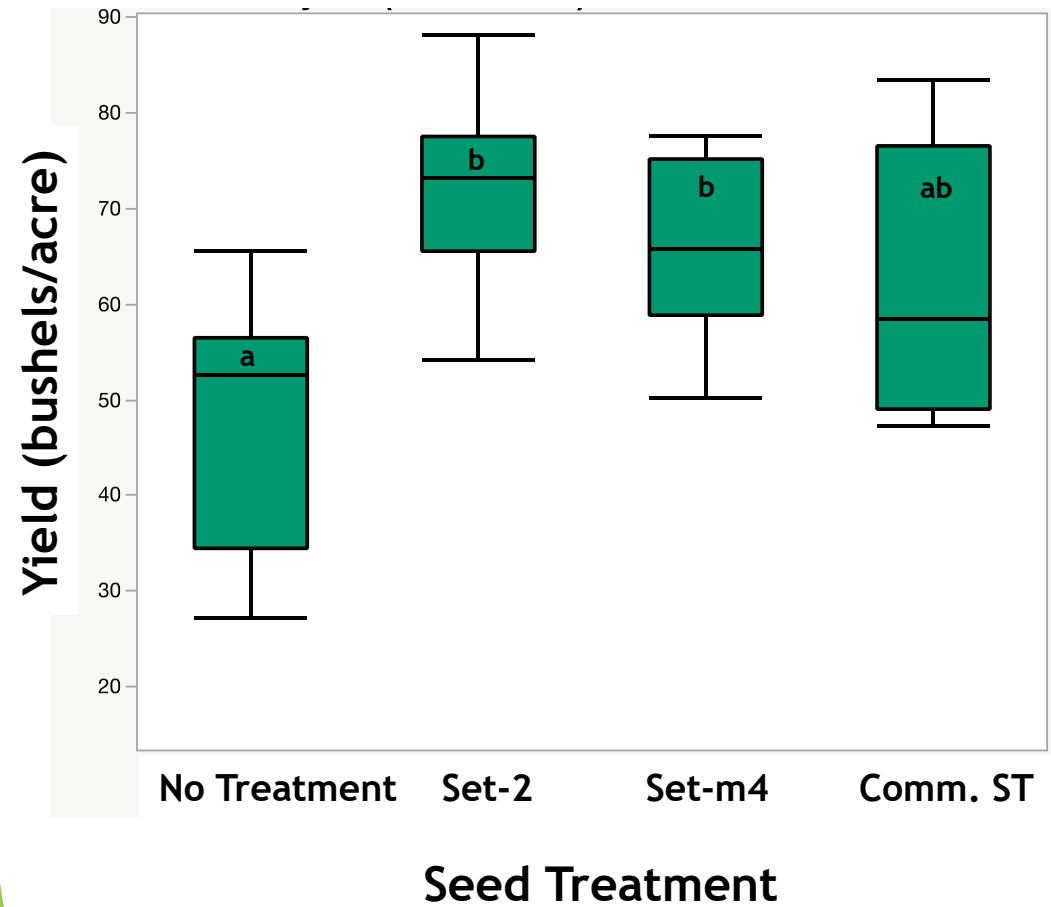
Doyle Chamber Research Station (Yield)



Dean Lee Research Station (Yield)

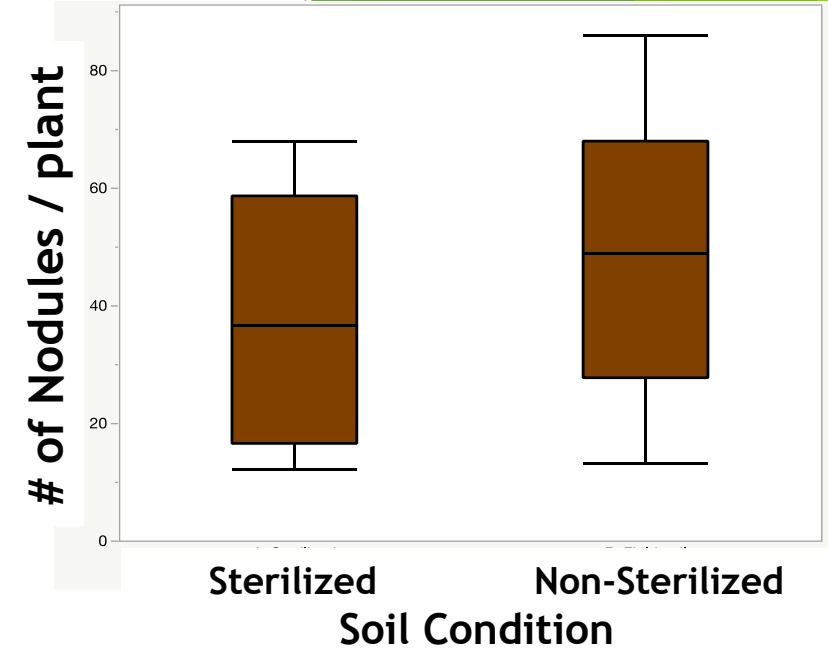


Red River Research Station (Yield)

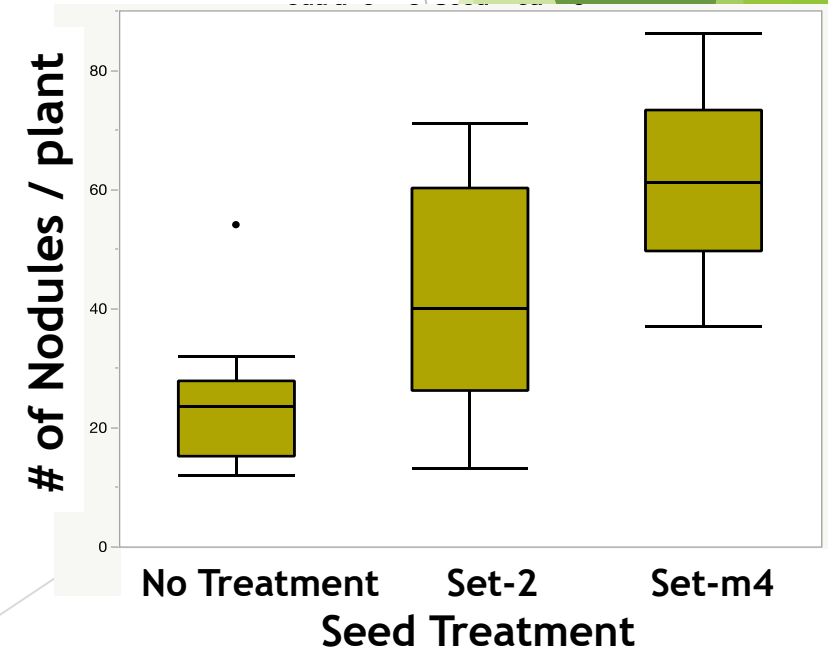


Effect of SABBs on Root Nodulation

**Sterilized
Soil Condition**



**Non-Sterilized
Soil Condition**



Development of Preservation Reagents

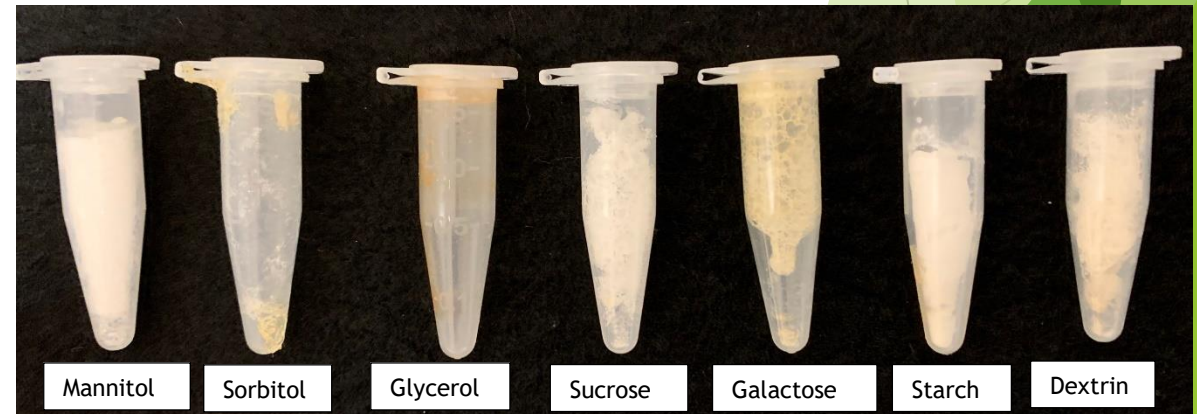
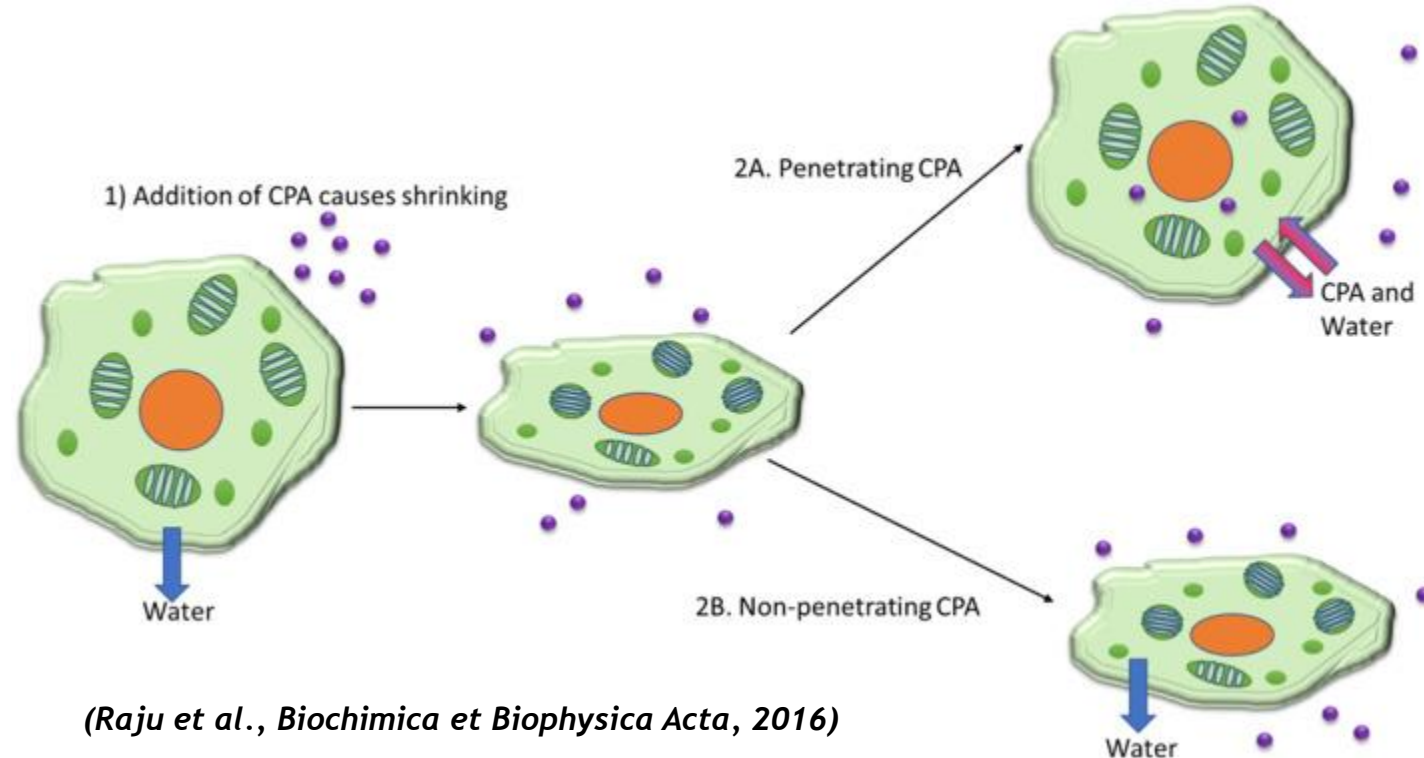
Types of cryoprotectants

- **Extracellular cryoprotectants**

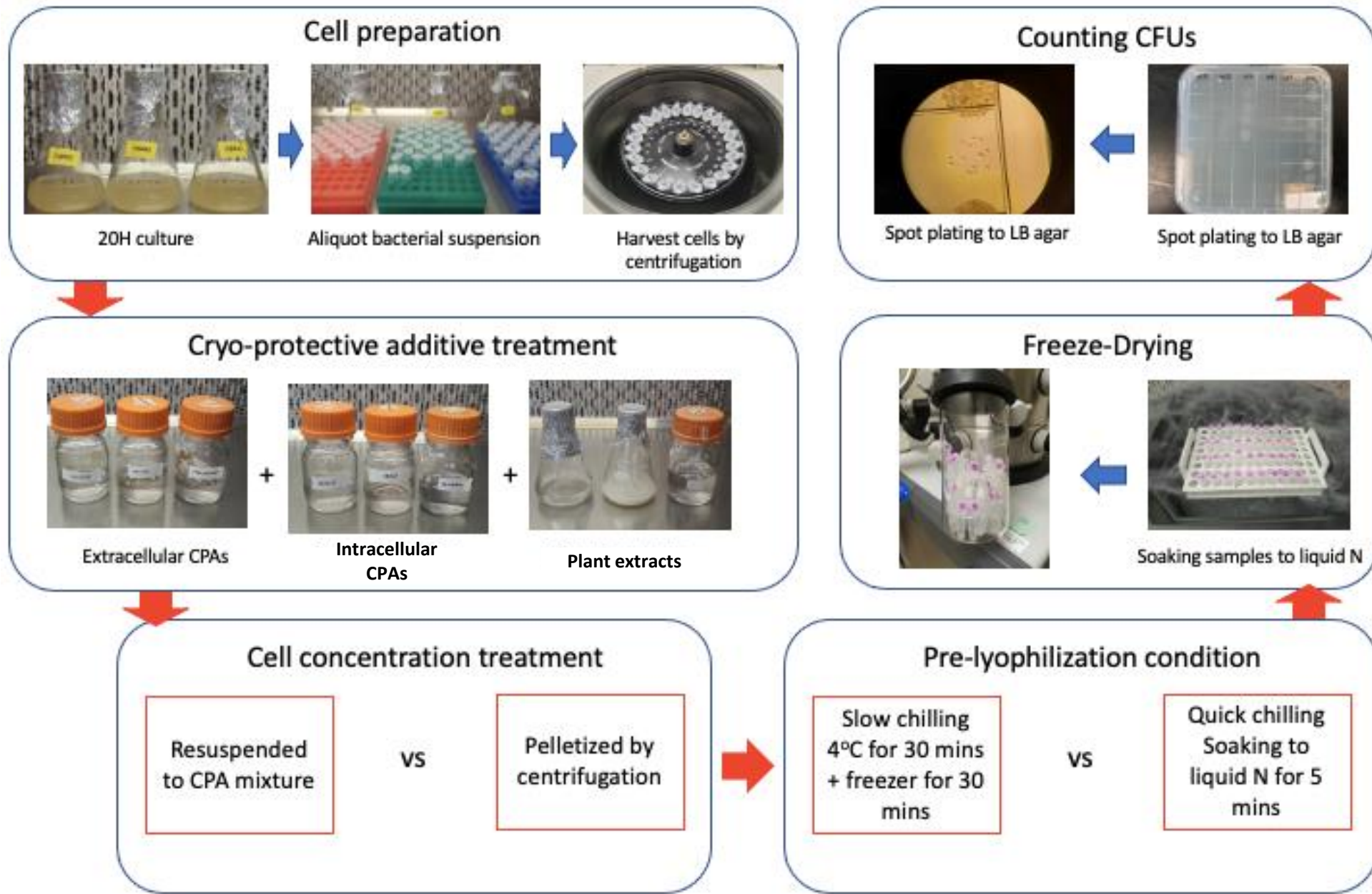
- ✓ Do not penetrate the bacterial cell wall
- ✓ Reduce the hyperosmotic gradients
- ✓ Include sucrose, cellobiose, polyvinylpyrrolidone (PVP), etc.

- **Intracellular cryoprotectants**

- ✓ Can penetrate the bacterial cell wall
- ✓ Reduce the water crystal formation within the cell
- ✓ Include DMSO, glycine, betaine, glycerol, etc.

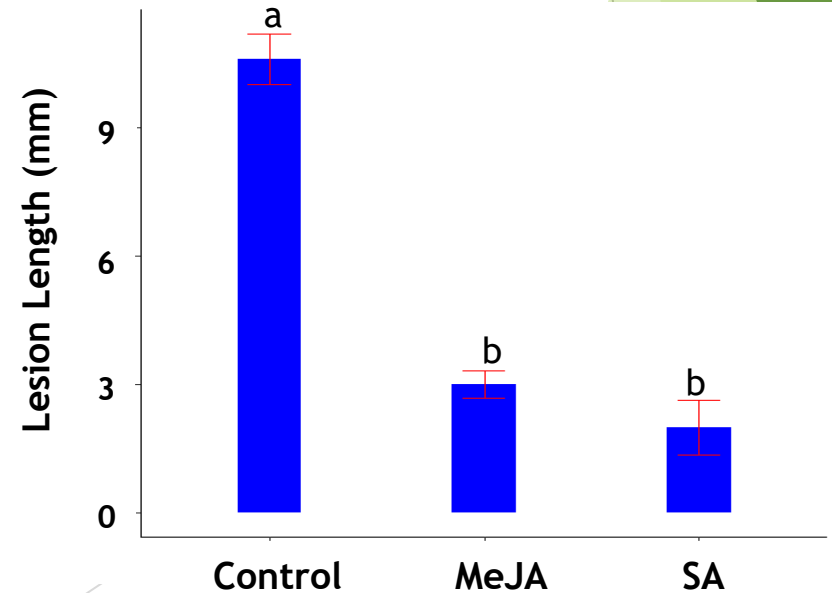
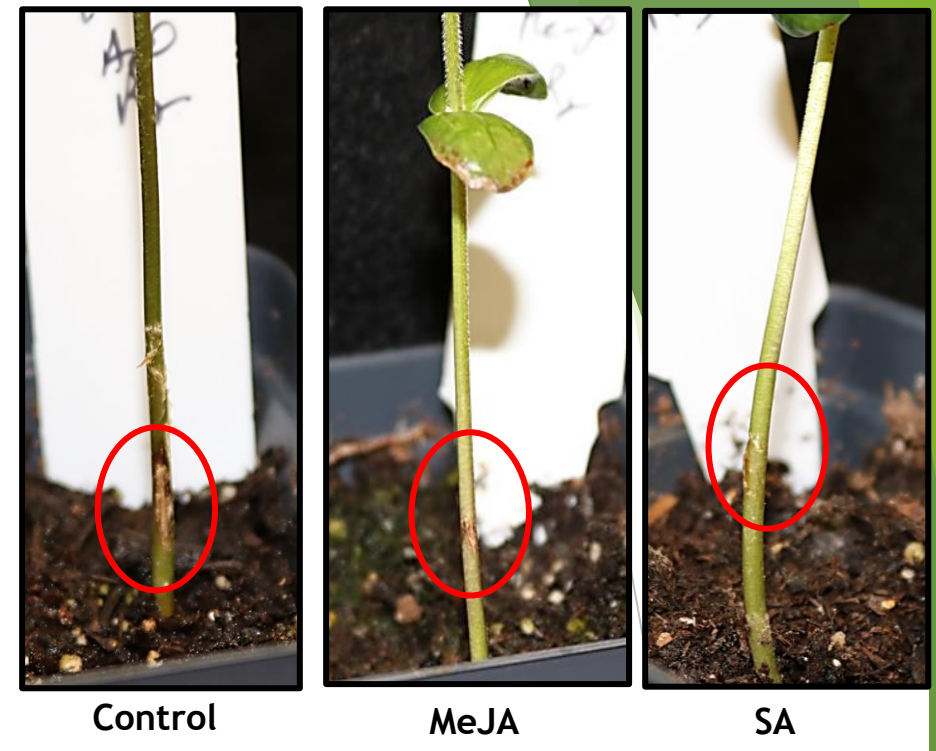


Optimization of bacterial cell lyophilization



Other Materials to Enhance Disease Resistance (Defense-Priming)

- If 'defense-priming' can be achieved through seed treatment, this practice to protect crops could be performed in more efficient and controlled ways.
- We recently observed that treatment of soybean seeds with salicylic acid or methyl-jasmonic acid caused enhanced resistance of soybean seedlings to the fungal pathogen, *Rhizoctonia solani*.
- Currently, we are testing additional materials to identify good defense-priming materials used for seed treatment (e.g. chitosan, BABA, VOCs, etc.)



Summary

- ▶ Soybean-associated beneficial bacteria (SABBs) were screened based on various biological activities beneficial to soybean growth.
- ▶ Bacterial mixtures (consortia) of SABBs having multiple beneficial activities showed higher growth-promoting activities than a single SABB through seed treatment, including increased yield and disease suppression as well as enhanced nodule formation.
- ▶ Currently, we are developing methodology and reagents for long-term storage of bacterial mixtures, using freeze-drying and various intracellular and extracellular cryoprotectants in combination with plant extracts.
- ▶ We are also studying candidate seed-treating materials for enhancing disease resistance of soybean (and rice)



Acknowledgements



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Soybean & Grain
RESEARCH & PROMOTION BOARD



Questions:

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