

Promotive Effects of  
*Paraburkholderia aspalathi*  
Rhizobacteria on Drought Stress  
Tolerance and Post-Drought  
Recovery

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Improving drought stress tolerance and post-drought recovery in cool season turfgrass is an important objective for reducing water use, while maintaining turf quality



## Drought Stress

- Reduced turf quality
- Chlorosis, yellowing
- Reduced growth and tillering
- Decline in canopy density
- Ethylene increases

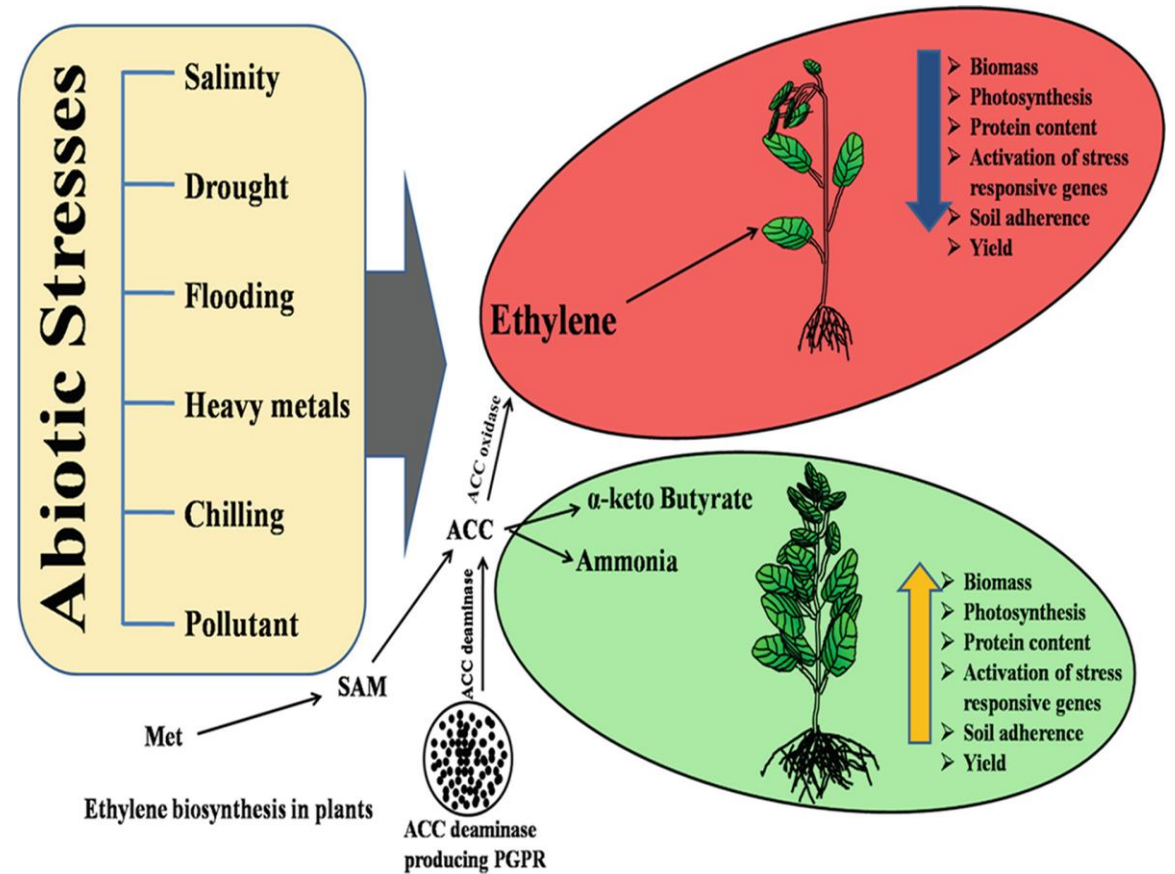
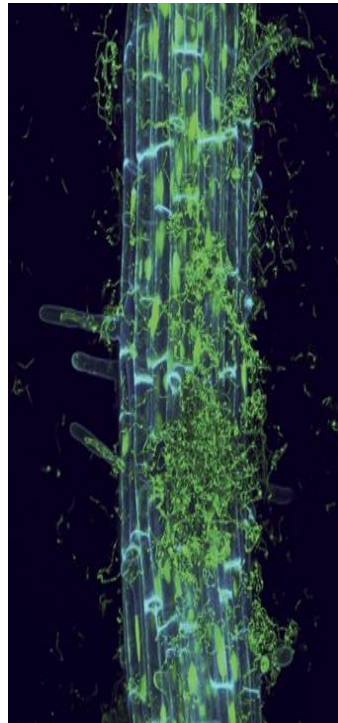


## Recovery

- Turf quality increases
- Ethylene levels are reduced
- Root viability for water and nutrient uptake
- Formation of new tillers
- Increased canopy density

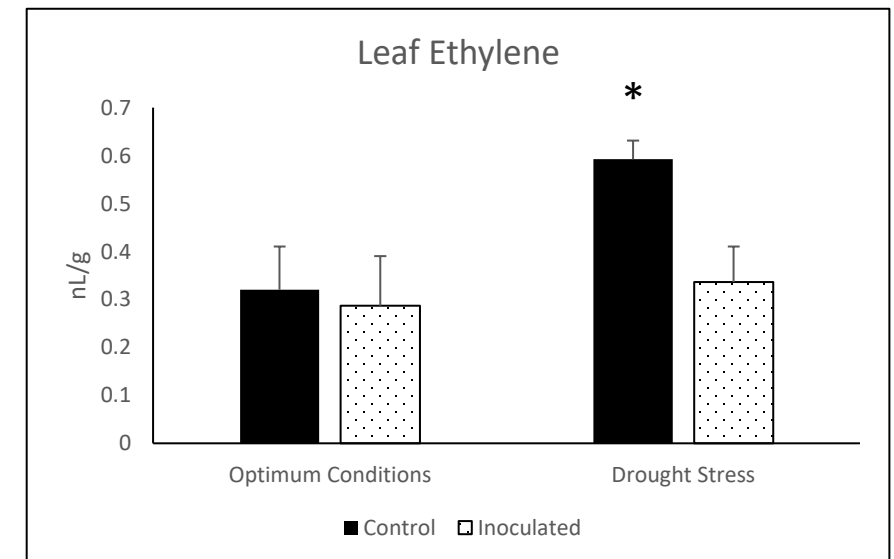
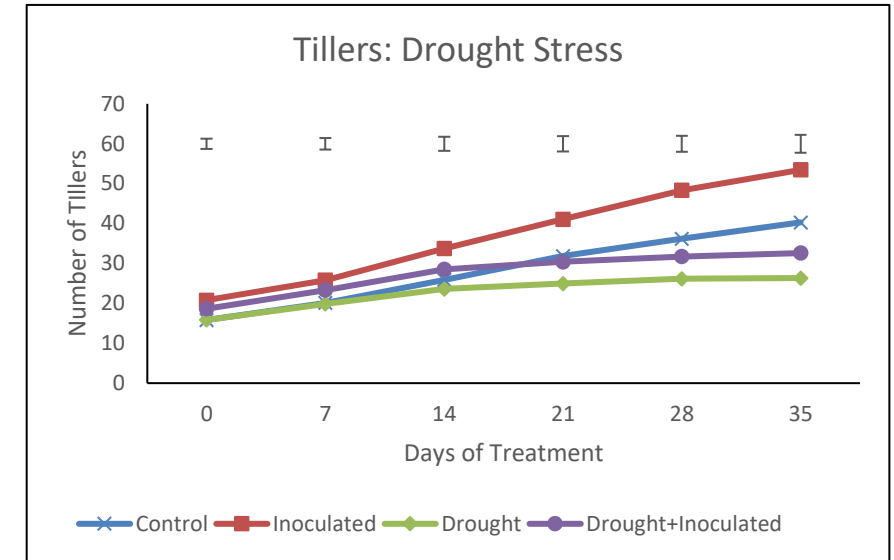
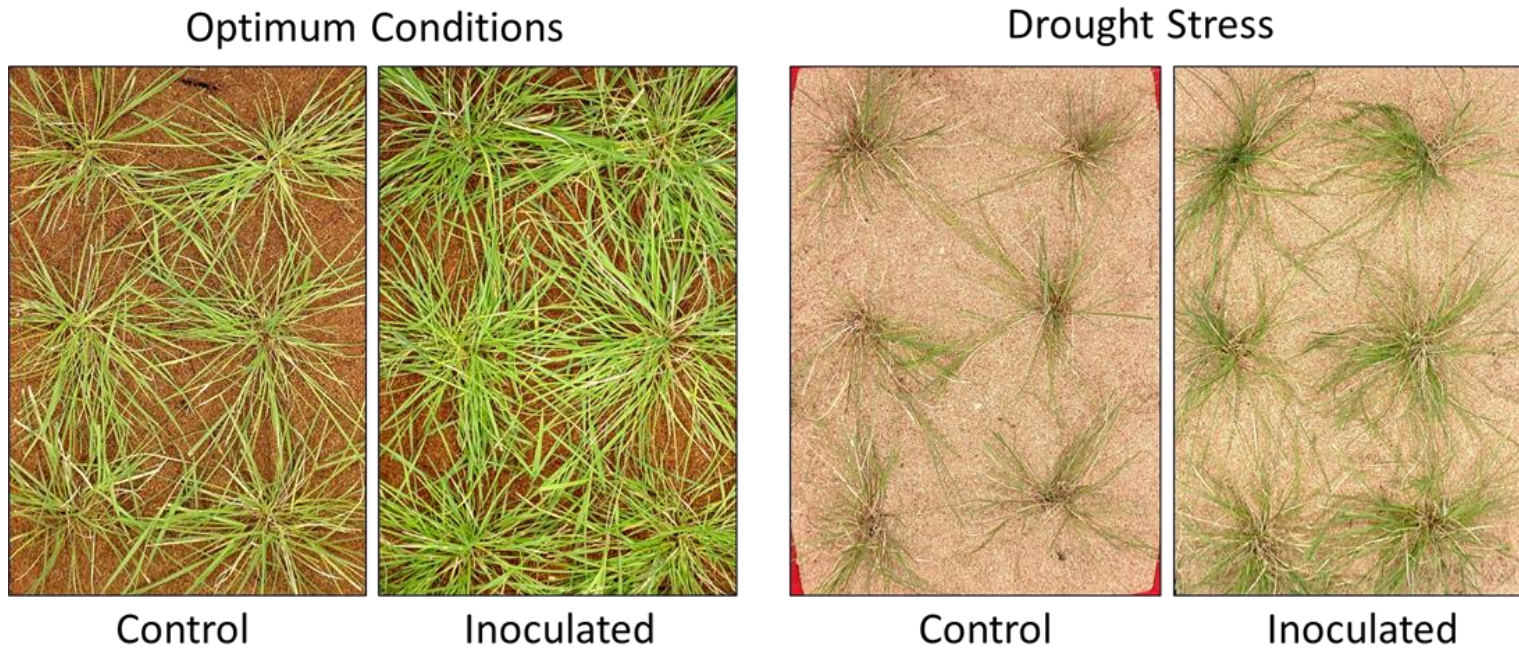
# Suppressing Ethylene Production by ACC Deaminase Producing Bacteria may Improve Drought Tolerance

- 1-Aminocyclopropane-1-carboxylic acid (**ACC**) – precursor of ethylene.
- Plant Growth Promoting Rhizobacteria (PGPR) with **ACC Deaminase (ACCd)** enzyme break down ACC into ammonia and  $\alpha$ -keto butyrate before ACC becomes ethylene.
- ACCd rhizobacteria utilize the nitrogen from ACC while plant roots benefit from the reduction in ethylene production.
- Reduced ACC  $\rightarrow$  Reduced Ethylene  $\rightarrow$  Reduced Stress Damage



# ACCd bacteria *Paraburkholderia aspalathi* enhanced tiller production by reducing ethylene concentrations during drought stress in creeping bentgrass

(Errickson and Huang, 2021 unpublished)



Success in controlled environment growth chambers can be challenging to replicate in field trials because variable environmental conditions and native soil organisms may influence the successful inoculation and effects of PGPR.

# Research Questions

Can these novel strains of *P. aspalathi* improve drought tolerance and post-drought recovery of cool season turfgrass in field conditions?

What is the most effective method and concentration for field inoculations of cool season turfgrass?

# Research Objectives

To evaluate physiological improvements in drought tolerance and post-drought recovery of cool season turfgrass inoculated with *P. aspalathi* in field conditions

To determine effective field inoculation methods and dosages for *P. aspalathi* in field conditions for cool season turfgrass

**Project 1**  
**Field Inoculation Methods**  
**(2020, 2021)**

**Project 2**  
**Field Dosage Optimization**  
**(2022)**



# Project 1: Field Inoculation Methods (2020, 2021)

- Fairway creeping bentgrass cv. Penncross field plots (1m x 1.3m, 4 replicates) maintained at fairway height.
- *P. aspalathi* strains 'WSF23' and 'WSF14' were applied at a concentration of  $1.0 \times 10^7$  CFUs in a 0.01% humic acid solution with a carrier volume of 2.0 gallons per 1,000 square feet.

## Inoculation Treatments

1. Non-inoculated control
2. Foliar spray
3. Soil drench

## Irrigation Treatments

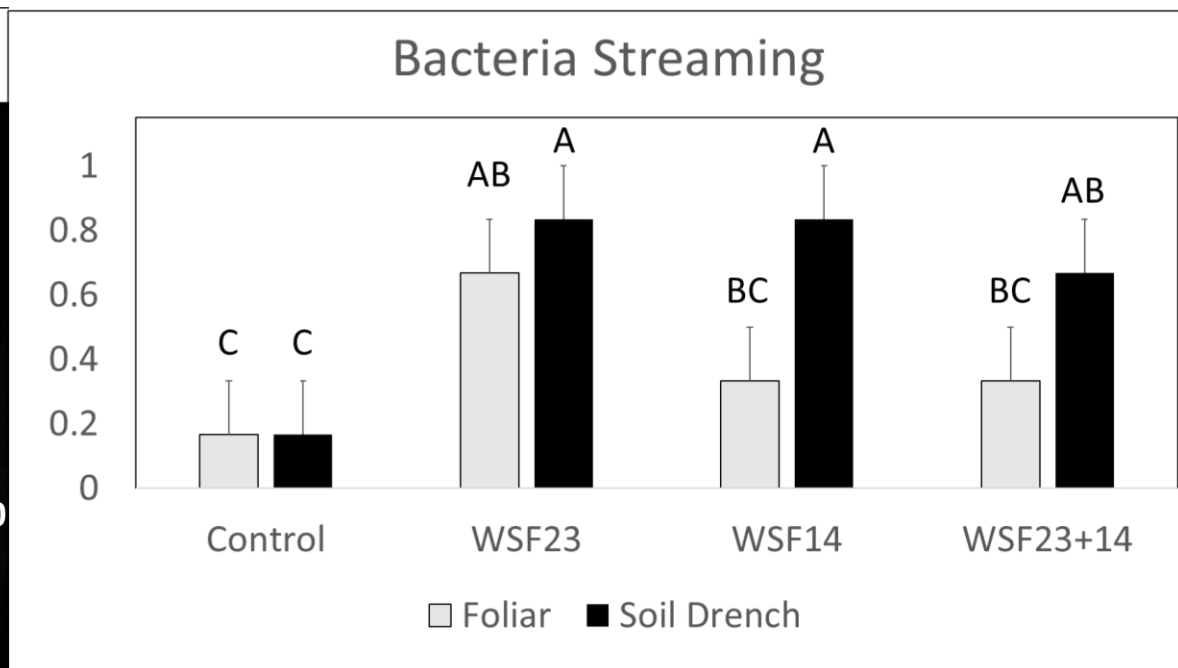
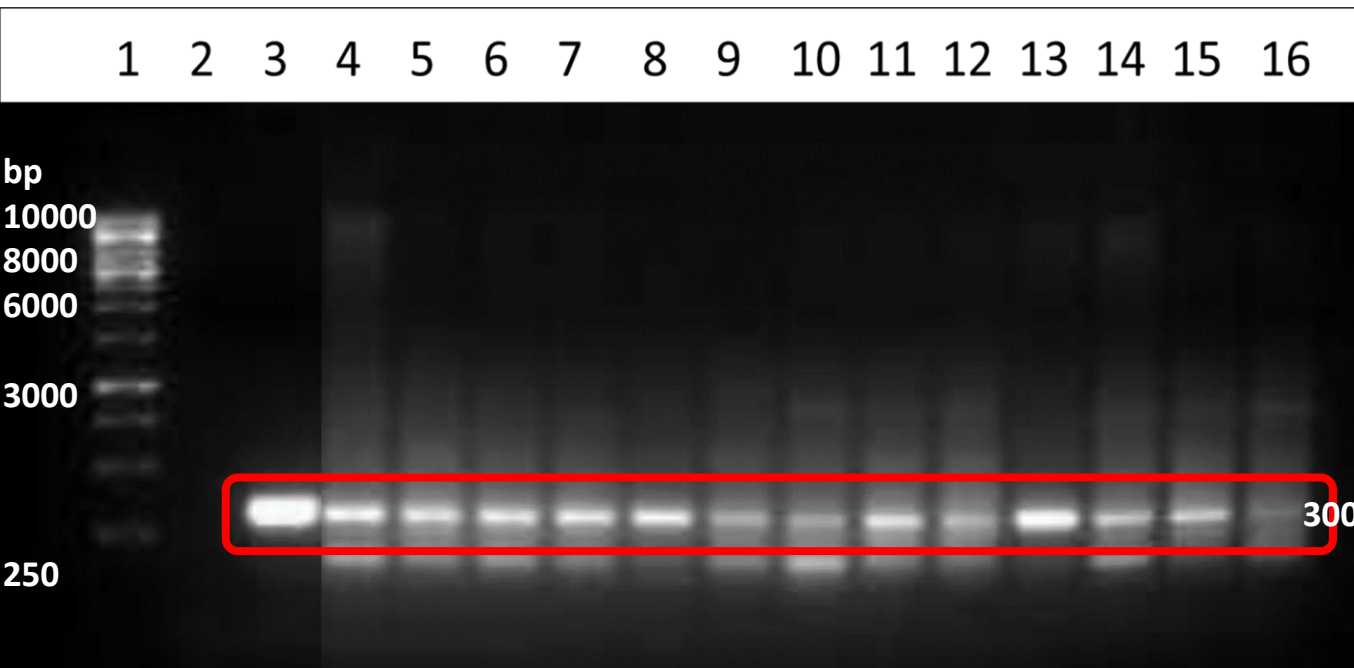
1. Well-Watered Control: Plants were irrigated (100% ET)
2. Drought Stress: Deficit Irrigation (60% ET) for 49 days (2020) and 28 days (2021)
3. Post-stress Recovery: Drought-stressed plants were re-watered for 28 days (2020) and 14 days (2021) (100% ET)



Weekly measurements of Turf Quality, NDVI, and Percent Green Cover



# Inoculation was confirmed using PCR analysis and bacterial streaming observations



Turf inoculated using the soil drench method had higher levels of bacterial streaming

The soil drench inoculation method was more effective than foliar application for promoting drought tolerance and post-drought recovery

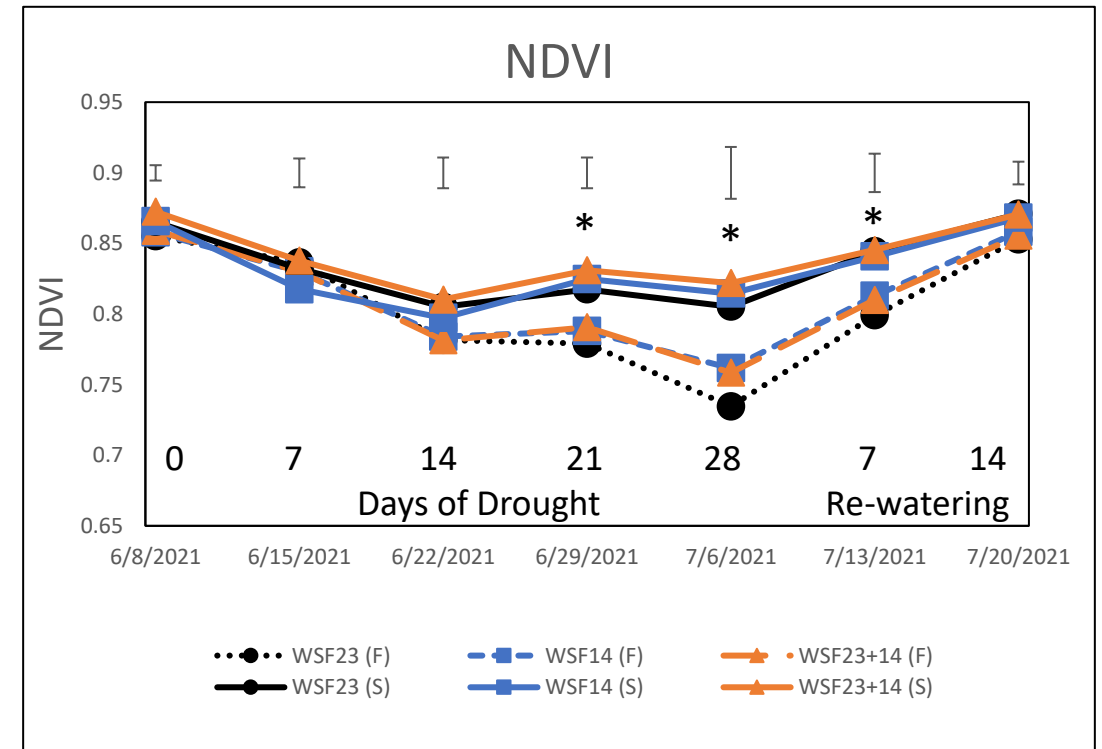
28 Days of Drought Stress (60% ET)



Control

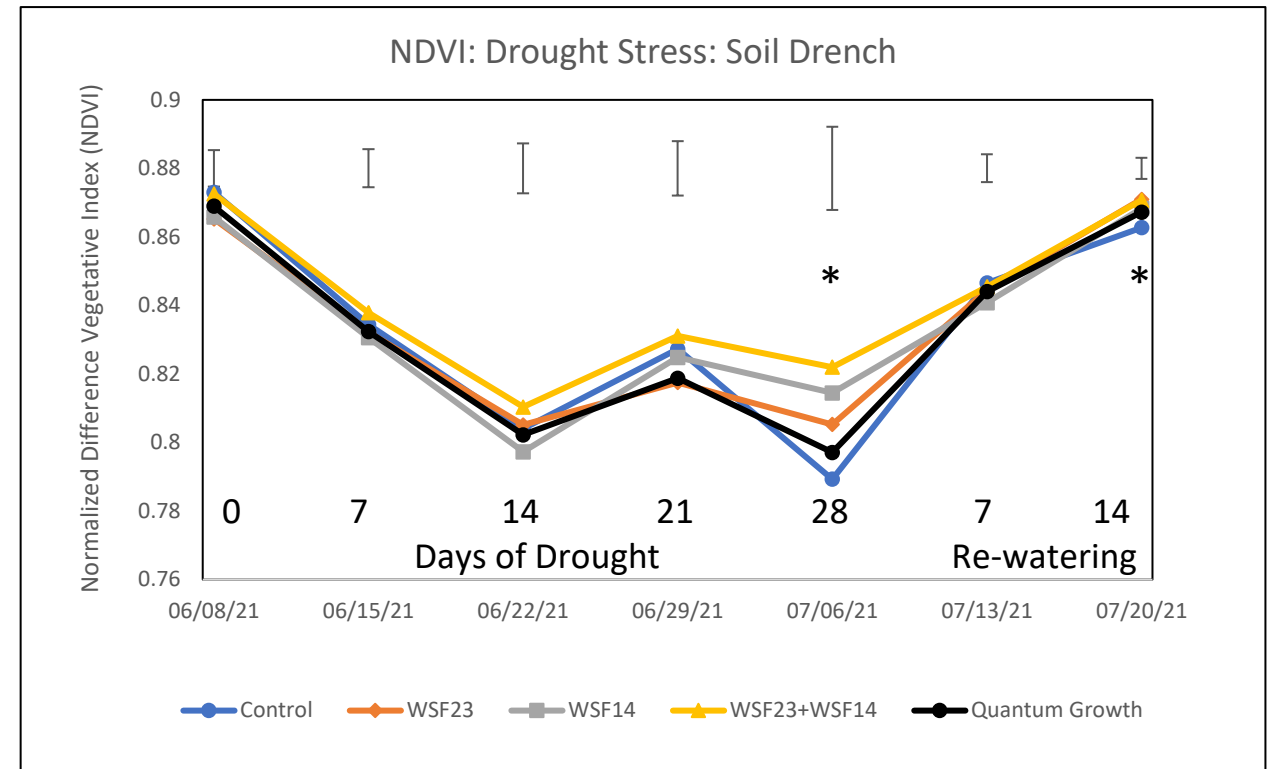
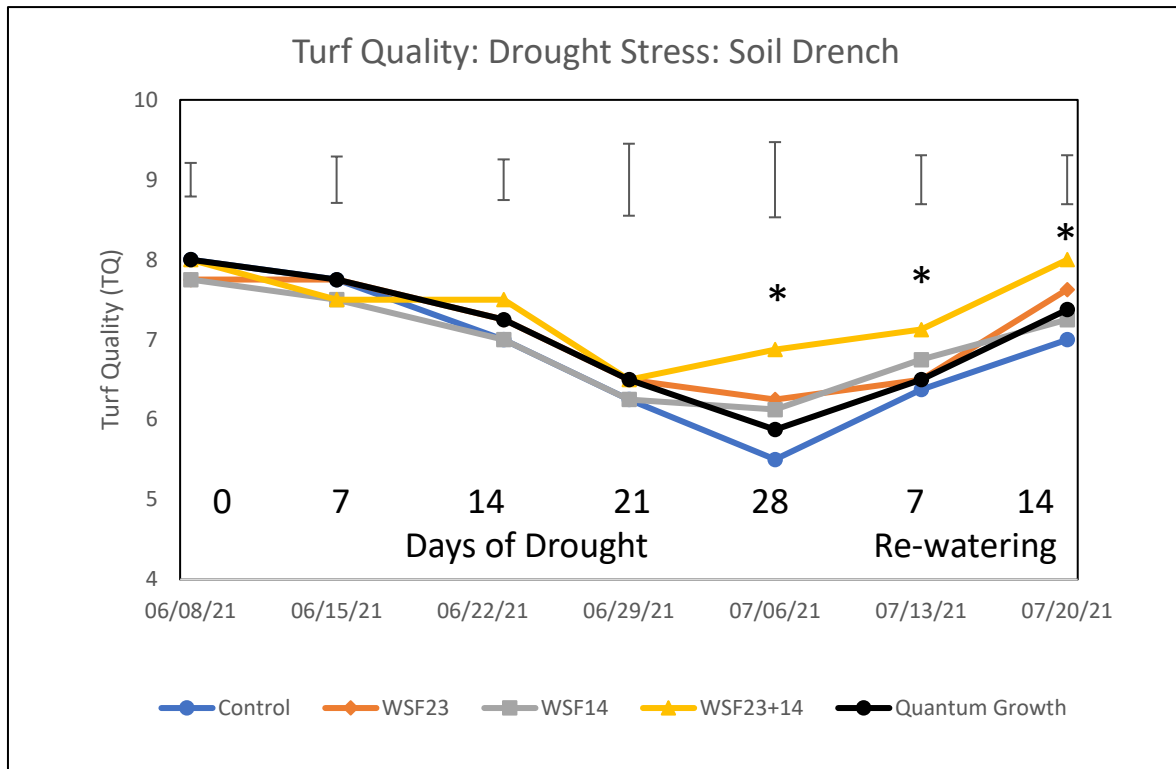
Foliar

Soil Drench



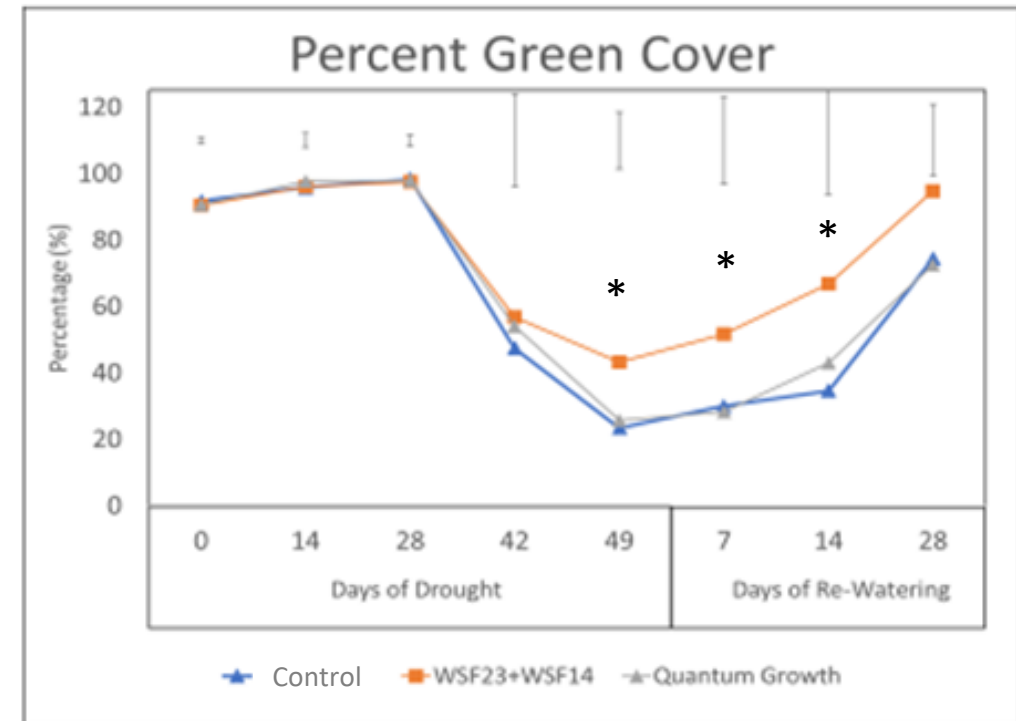
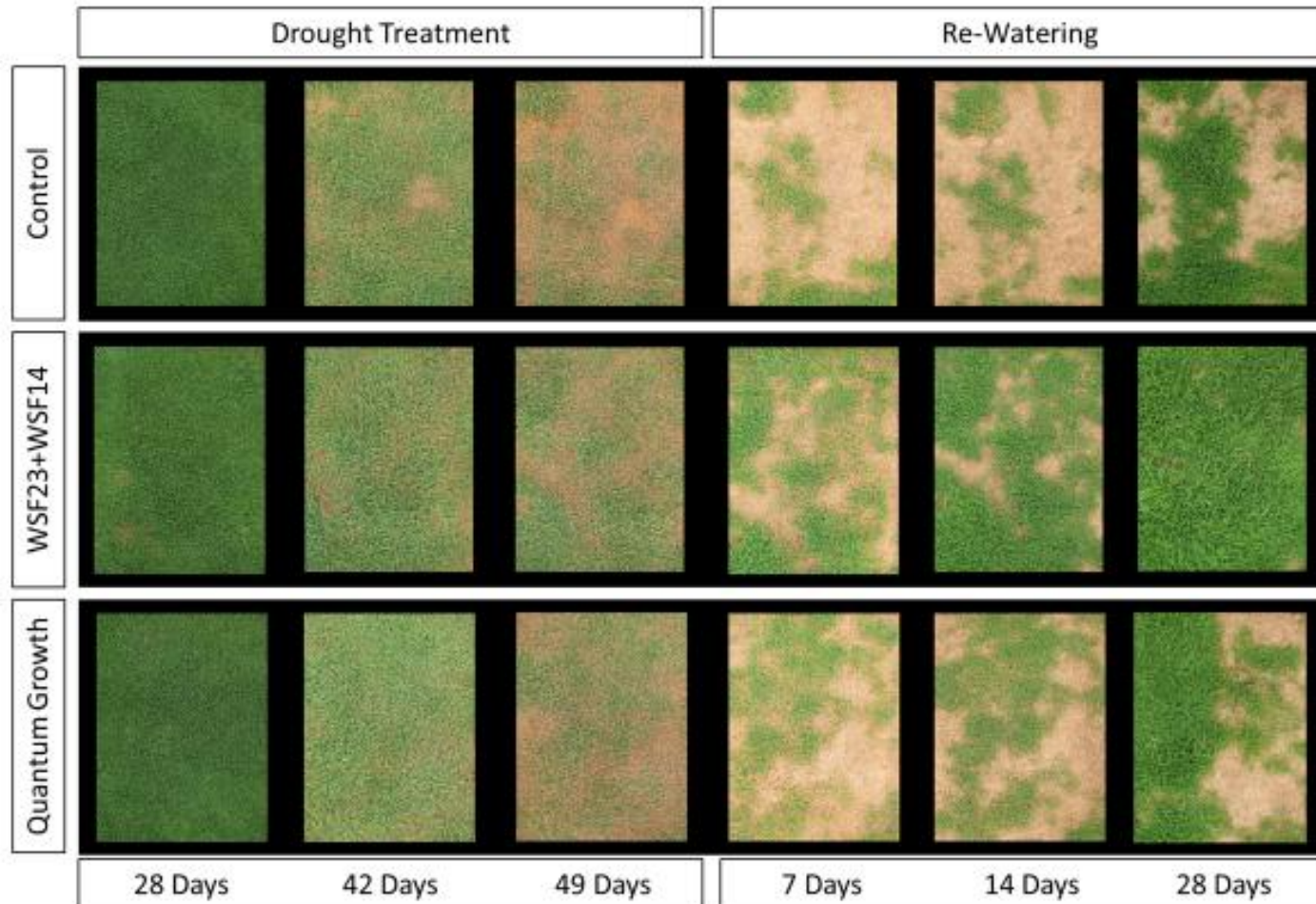
\* Indicates significance at  $p < 0.05$

The combination of PGPR strains WSF23 and WSF14 was more effective than each strain individually for improving turf quality and NDVI during drought stress and post drought recovery



\* Indicates significance at  $p < 0.05$

Canopy density was highest in turf inoculated with a combination of WSF23 and WSF14 using the soil drench method



\* Indicates significance at  $p < 0.05$

# Project 2: Field Dosage Optimization (2022)

- Fairway creeping bentgrass cv. Penncross field plots (1m x 1.3m, 4 replicates) maintained at fairway height.
- *P. aspalathi* strains 'WSF23' and 'WSF14' were applied at varying rates using the soil drench method.

## Inoculation Treatments

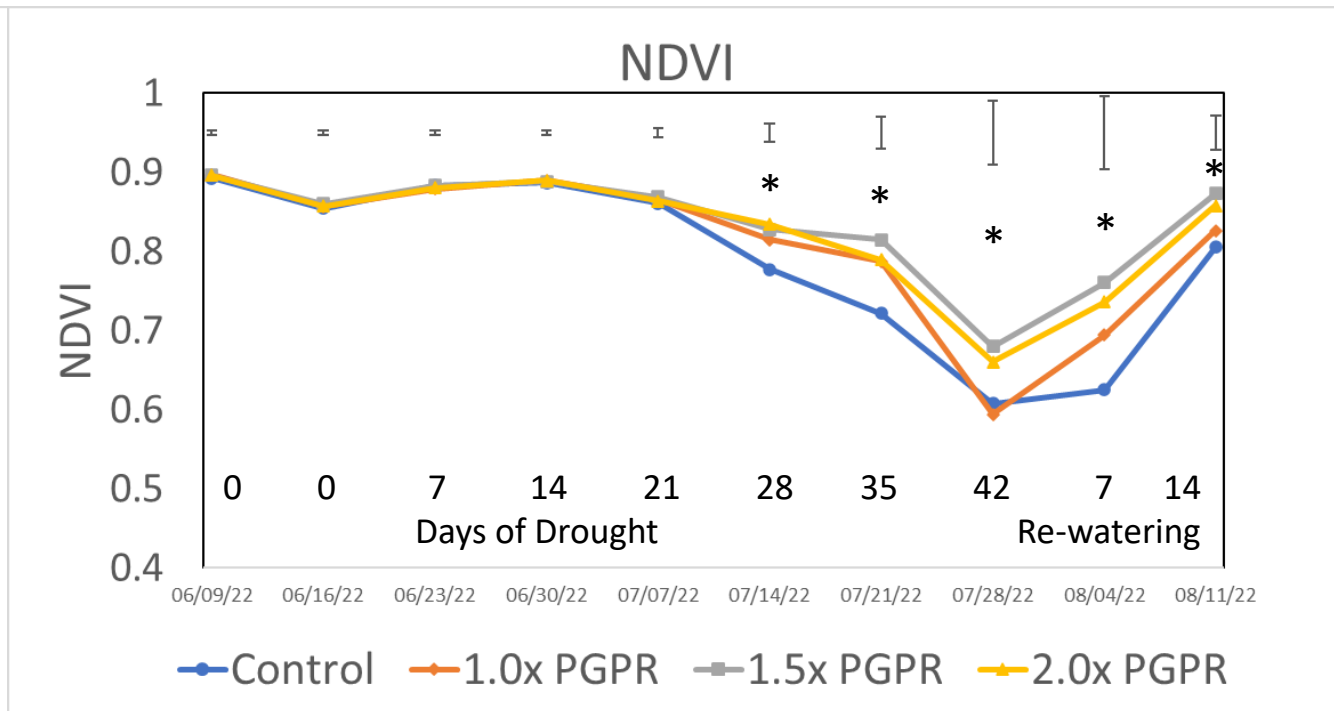
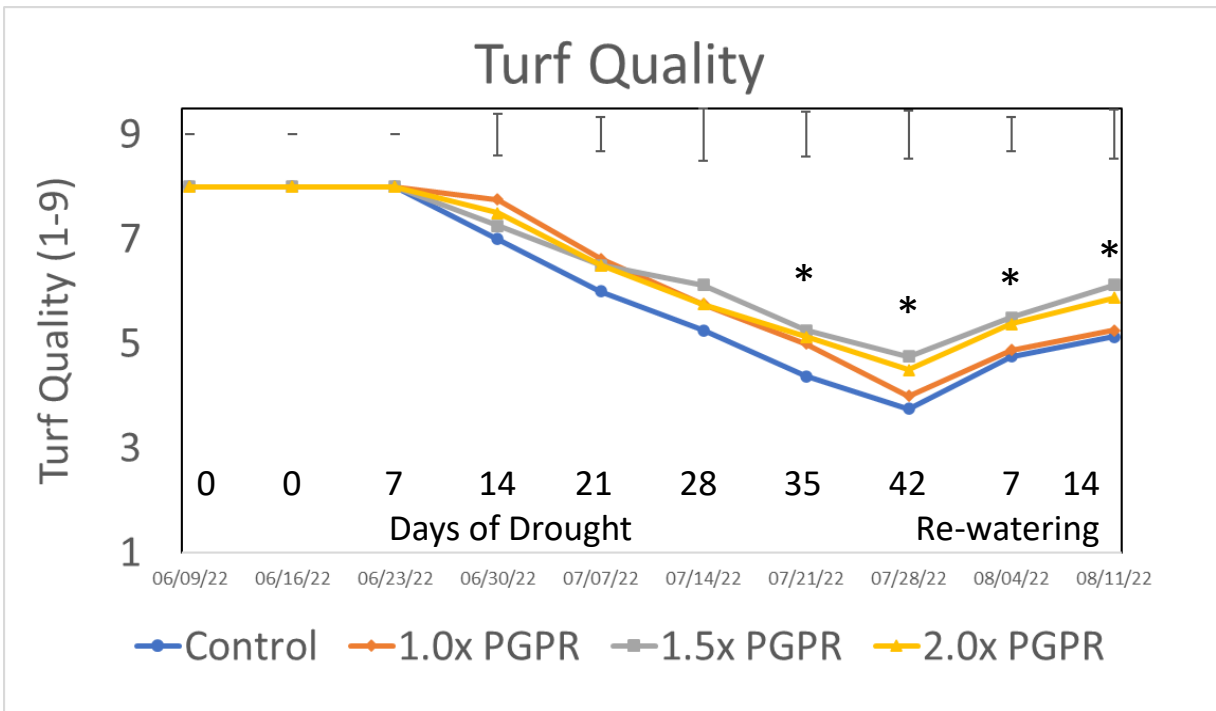
1. Non-inoculated control
2.  $1.0 \times 10^7$  CFUs
3.  $1.5 \times 10^7$  CFUs
4.  $2.0 \times 10^7$  CFUs

## Irrigation Treatments

1. Drought Stress: Deficit Irrigation (60% ET) for 35 days
2. Post-stress Recovery: Drought-stressed plants were re-watered for 14 days (100% ET)



# Inoculation with the 1.5x PGPR concentration had the greatest improvement on Turf Quality and NDVI



\* Indicates significance at p<0.05

**Project 1**  
**Field Inoculation Methods**  
**(2020, 2021)**

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graph LR; P1[Project 1  
Field Inoculation Methods  
(2020, 2021)] --> R1[Soil drenching was more  
effective for rhizobacteria  
colonization and improving  
drought tolerance and post-  
stress recovery]; P2[Project 2  
Field Dosage Optimization  
(2022)] --> R2[Higher rate or concentration  
of rhizobacteria was more  
effective for field inoculation];
```

**Soil drenching was more effective for rhizobacteria colonization and improving drought tolerance and post-stress recovery**

**Project 2**  
**Field Dosage Optimization**  
**(2022)**

**Higher rate or concentration of rhizobacteria was more effective for field inoculation**

Inoculation with a combination of two novel strains of PGPR (*P. aspalathi* 'WSF23' and 'WSF14') using the soil drench method was effective at improving drought tolerance and post-drought recovery of cool season turfgrass under field conditions

- Fairway creeping bentgrass demonstrated increased turf quality, NDVI, and canopy density with inoculation
- Increased plant density and lateral spread due to stimulated tiller production
- Related to suppression of stress-induced ethylene



# Significance and Future Work

- *P. aspalathi* 'WSF23' and 'WSF14' have potential as commercial inoculants for turfgrass managers to reduce water use.
- Future work will investigate the metabolic and physiological mechanisms of how rhizobacteria *P. aspalathi* 'WSF23' and 'WSF14' improve drought tolerance and the interaction of ethylene suppression with other hormones controlling tiller production.

# Thank You

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- Huang Lab Members
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- USDA SARE

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