

Utilizing Microbial Endophytes to Inhibit Germination of King Ranch Bluestem (*Bothriochloa ischaemum* var. *songarica*): A Novel Tactic for Rangeland Restoration

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Introduction:

- King Ranch Bluestem (*Bothriochloa ischaemum* var. *songarica*) is a perennial bunchgrass from the family Poaceae.
- Introduced to Texas in 1939 as a forage crop for grazing and used for pasture, hay production, and erosion control.
- King Ranch Bluestem is now spreading across the Southern US
- It can outcompete native grasses, reducing plant diversity and the variety of habitats for wildlife.
- Endophytes are microorganisms (fungi or bacteria) that live inside plant tissues without causing harm.
- The goal is to identify native endophytes that help promote the growth of native grasses while reducing the spread of King Ranch bluestem.

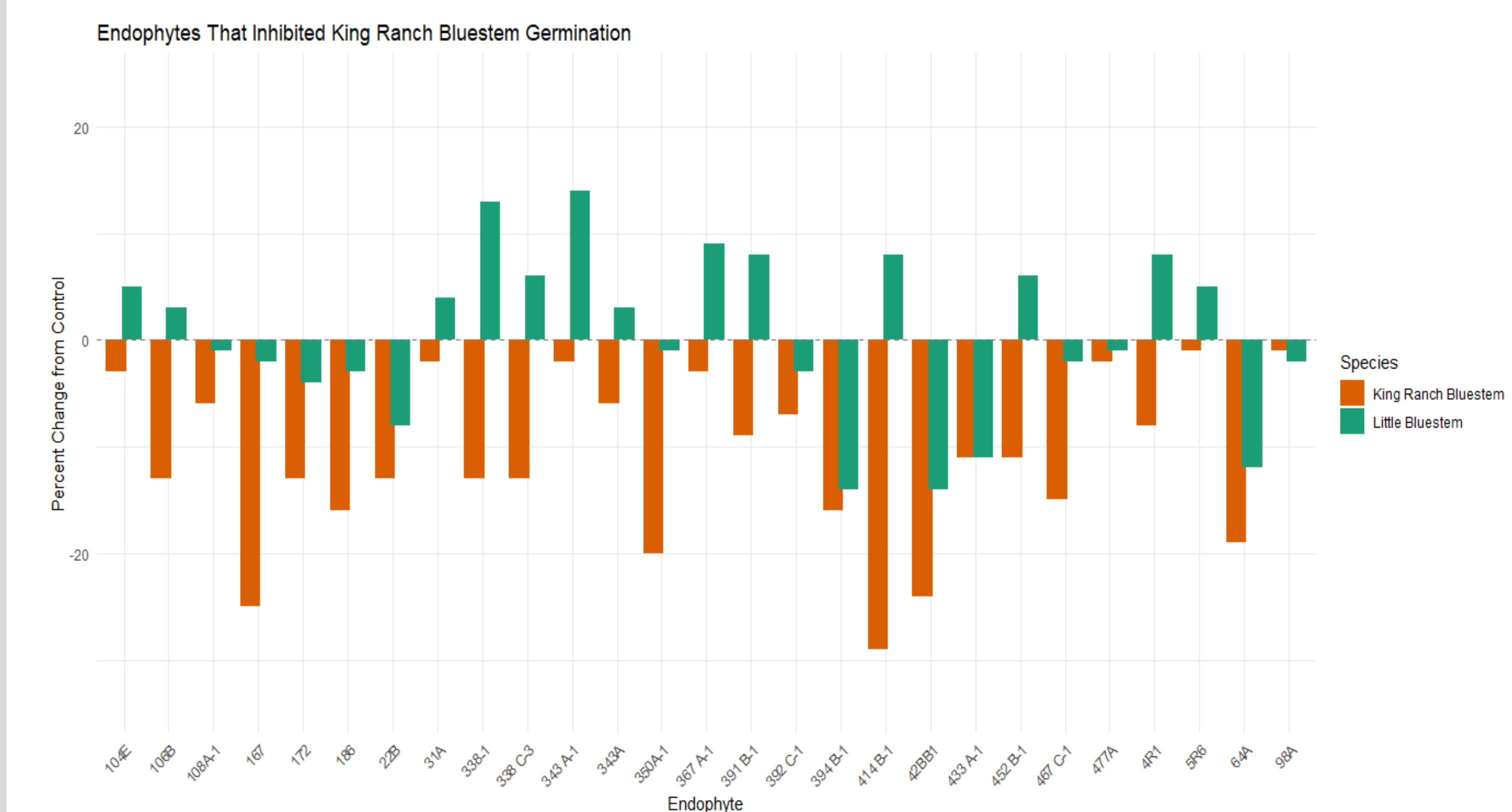
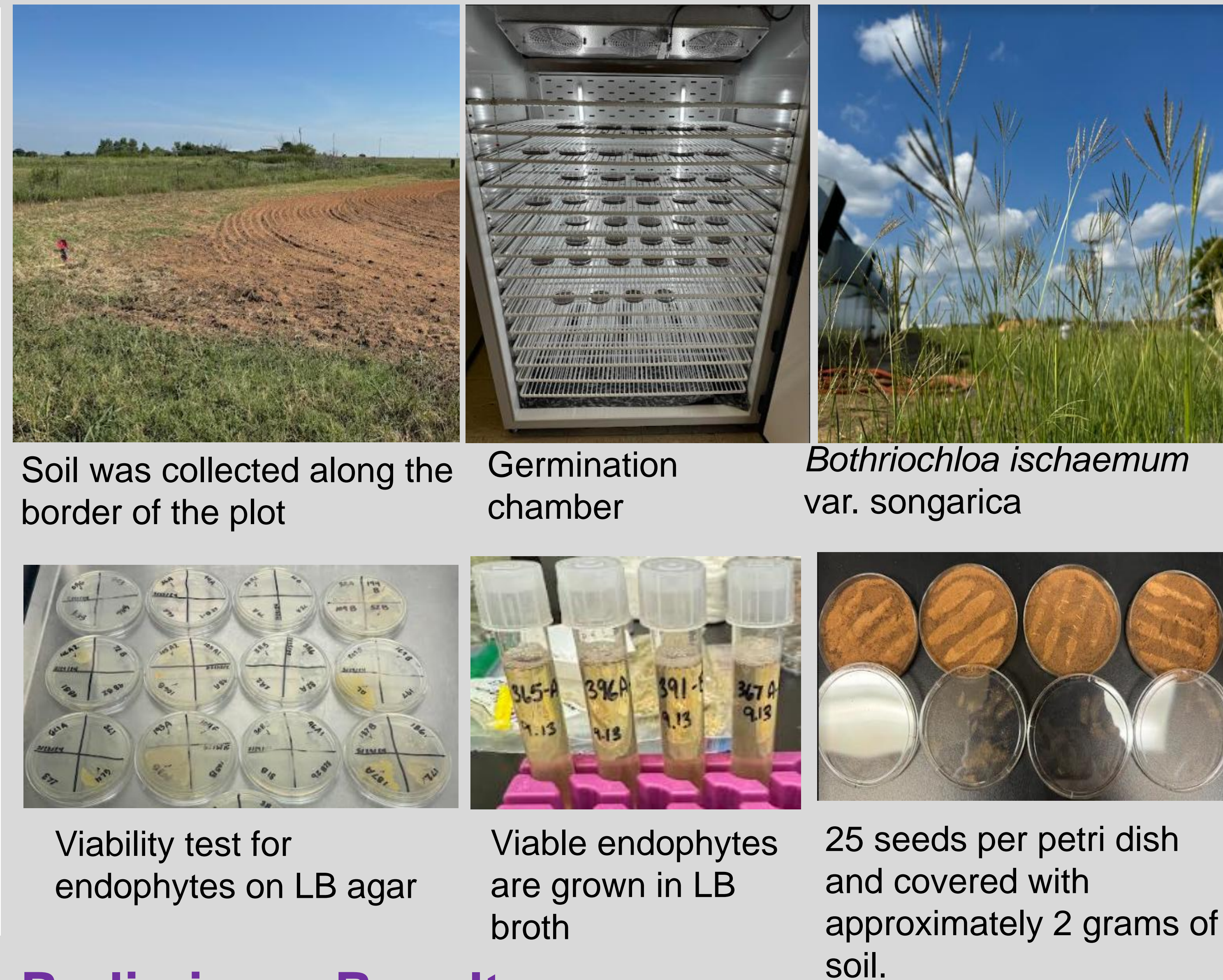


Figure 1: Endophyte treatments that decreased germination in King Ranch bluestem with corresponding effects on Little bluestem.

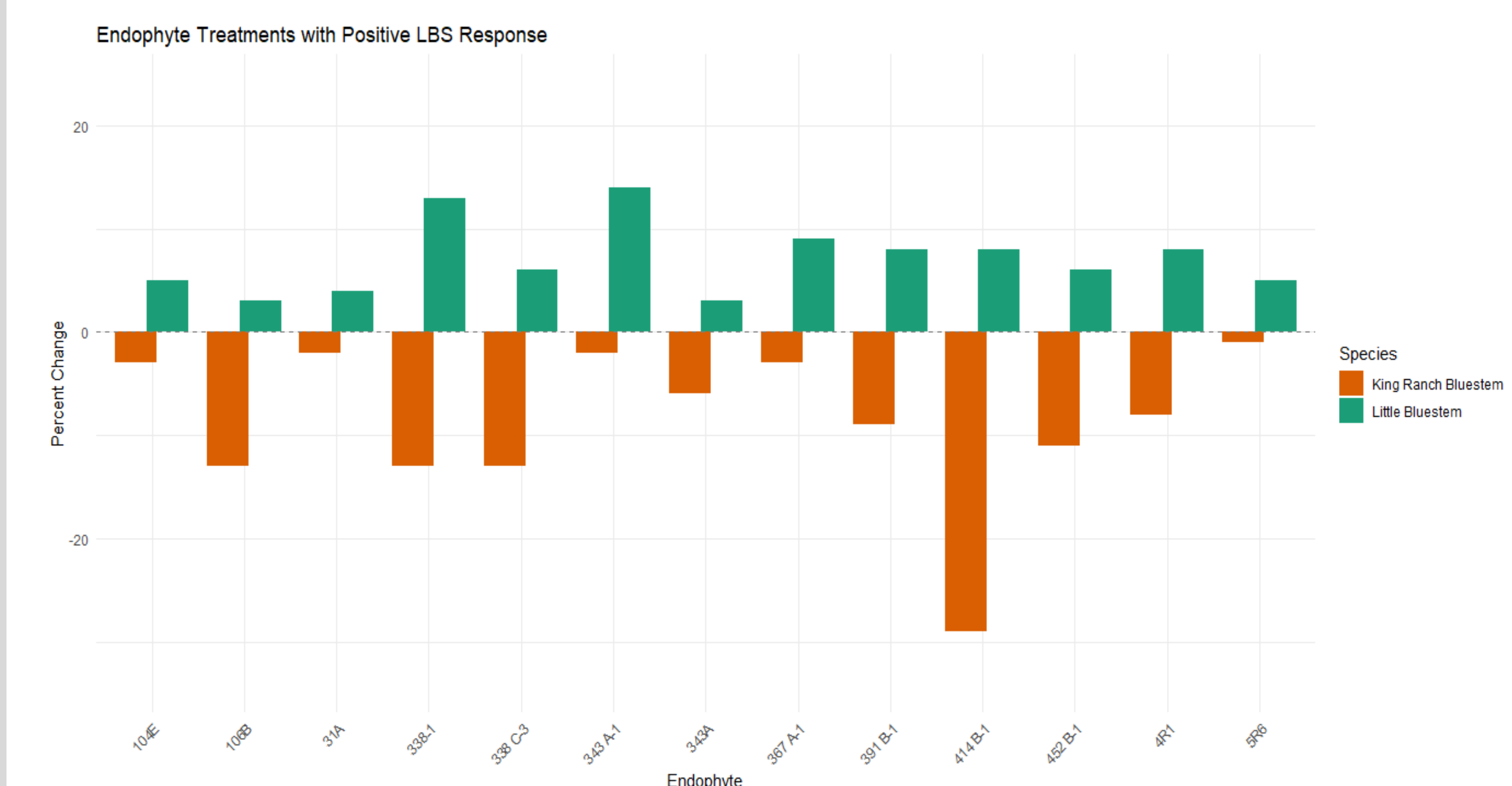


Figure 2: Endophyte treatments that increased germination in Little bluestem, with corresponding effects on King Ranch bluestem.

Methods:

Testing for Viability:

- Endophytes from 2021 native grass microbial collection are streaked on LB agar

Seed and Soil Collection/Preparation:

- Seeds and soil were collected from the border of the AgriLife Center at Stephenville

Prepare Microbial Inoculant:

- Streak endophytes from glycerol stock onto LB agar plates and incubate.
- Transfer colony to a culture tube with 10 mL of 1X LB broth; incubate for 1-2 days.
- Centrifuge and resuspend pellet in 10 mL of 0.1X LB broth.
- OD reading, serial dilutions for PCR, and CFU counts.

Germination Chamber Setup:

- Program chamber for day/night cycles (Day: 26°C, 66% RH; Night: 20°C, 50% RH).

Statistical Methods:

- ANOVA for factorial, repeated measures analysis and Tukey's multiple mean comparisons.
- Germination rates, growth parameters, and biomass

Preliminary Results:

Table 1: CFU Counts and OD₆₀₀ Readings for endophytes screened. Colony forming units (CFUs) were counted from samples diluted to 10⁻⁵. The OD₆₀₀ readings were taken before the sample was diluted.

Endophyte	Coloines Counted	CFU counts (CFU/mL)	OD ₆₀₀
350A-1	47	2.35 × 10 ⁷	0.258A
167	153	7.65 × 10 ⁸	0.640A
414 B-1	226	1.13 × 10 ⁷	0.210A
343 A-1	245	2.05 × 10 ⁸	0.202A



King Ranch Bluestem seeds removed from the soil (below) at the end of 14-day germination.



Little Bluestem seeds removed from the soil (below) at the end of 14-day germination.



References

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