Drought Stress Adaptation in Winter Wheat through Soil Microbial Interactions and Root Architecture

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Belowground interactions with ACC deaminase-positive bacteria

- Bacteria that degrade ACC, the precursor to stress ethylene

Methionine $\rightarrow$ S-adenosyl methionine

$\xrightarrow{\text{ACC SYNTHASE}}$

$\text{1-amino-cyclopropane-1-carboxylic acid}$

$\xrightarrow{\text{ACC OXIDASE}}$

$\text{ethylene}$

$\xrightarrow{\text{ACC DEAMINASE}}$

$\text{α-ketobutyrate}$
Preliminary findings

- ACC+ bacteria are abundant in CO soil
  - Field study at LIRF with 4 cultivars
- Abundance and bacterial species composition varies with different wheat varieties
  - Ripper vs. RonL cultivars
- Some varieties are more responsive to inoculation than others
  - Greater productivity of RonL vs. Ripper
  - Different root exudate chemicals?
Hypothesis and Objectives

H: Water uptake of wheat can be improved through interactions with specific ACC+ bacteria

Obj 1: Correlate winter wheat responses to water stress under field conditions with the genetic diversity and activity of ACC+ bacteria in the rhizosphere.

Obj 2: Identify variation among wheat genotypes in root exudate chemical profiles, and effects of ACC+ bacterial inoculation on root architecture and above-ground growth and water status, under water-stressed and non-stressed conditions.

Obj 3: Quantify the increase in drought tolerance of drought susceptible wheat cultivars grown in the presence of ACC+ bacteria and root exudates of drought tolerant cultivars.
Obj 1 update

Field study in 2013
- 12 cultivars at ARDEC
- Dryland or fully irrigated
- Rhizosphere soil collected at mid-grain filling
- ACC deaminase activity
- Bacterial diversity by 454 pyrosequencing
ACC deaminase activity is correlated with bacterial biomass.

\[ y = 0.8862 \ln(x) - 16.986 \]

\[ R^2 = 0.3101 \]

\[ P = 0.007 \]
Obj 2 update

- Greenhouse study complete
  - 7 cultivars
  - Inoculated with or without ACC+ bacteria
  - Grown with or without water stress
  - 4 replicates
  - Leaf relative water potential, root architecture, and root exudates
Greehouse study set-up

Root trait evaluation in a fritted clay growth medium allowing clean separation of the root mass.
WinRhizo analysis

Diameter (mm)
- 1.0+
- 0.75 – 1.0
- 0.50 – 0.75
- 0.25 – 0.50
- 0.0 – 0.25

Tolerant

Sensitive

0-33 cm

33-66 cm

66-99 cm
Total Root Length in 3 Tube Sections

Dry

Wet
Inoculation Effects

- Significant inoculation × cultivar effect on total root biomass at the bottom of the root tubes
  - Under water stress, RonL showed the largest increase (145%) in root biomass in response to inoculation

- Significant effect on leaf relative water content
Leaf relative water content

Inoculation effect $P = 0.03$

- Byrd
- Hatcher
- OK06318
- Ripper
- RonL
- Tam 12
- WB Cedar

ACC+ and Control
Metabolomics of Root Exudates

- Exudates from sterile sand columns that roots have grown into
- Extraction in water and methanol
- GC-MS and UPLC-MS-MS analysis at the Metabolomics and Proteomics Core Facility
Obj 3 update

- Greenhouse study is underway
  - 3 cultivars, grown as bi- or monocultures
    - 2 responsive to ACC+ bacteria (RonL, OK6318)
    - 1 non-responsive (Byrd)
  - Inoculated with or without ACC+ bacteria
  - Grown with or without water stress
  - 6 replicates
  - Yield traits at physiological maturity
RonL or OK6318 + Byrd → Byrd with improved drought tolerance

Byrd + Byrd → Byrd without improved drought tolerance
On-going Research and Goals

- USDA NIFA proposal
  - Plant Growth and Development, Composition and Stress Tolerance
- Identify root exudate(s) that recruit and enrich beneficial ACC+ bacteria
- Identify genetic marker(s) associated with root exudates and ACC+ bacteria
- Breed new drought-tolerant cultivars that combine multiple drought tolerance traits
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