Non-invasive Inoculation Method of Tomato Fruit with *Geotrichum candidum*

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Outline

- Introduction and Objective
- Proposed Vacuum Method
- Validation of Non-invasive Method
- Results & Comparison to Established Method
- Future Use and Testing
Tomato Sour Rot

- *Geotrichum candidum* is the causal agent of sour rot in tomato fruits
- Saprophytic “yeast-like” organism
- Major losses in-field and particularly postharvest
- Disease can proliferate in cold room storage conditions (55°F)
- Larger outbreaks are seen as a result of fall harvests characterized by significant rainfall events, heavy fogs, and fluctuating temperatures.
Sour Rot Control and Suppression

- Currently there are no in-field treatments to control sour rot infections
- Limited postharvest treatments
  - Harvest when plants and fruit are dry
  - Follow correct postharvest sanitation protocols
  - Use of propiconazole (Expected label in near future)
    - Effective when tested with wounding method
- Research needed to better understand pathosystem in hopes of improving disease control
Past Screening Methodology for Efficacy on Tomato Sour Rot

- Punch 4- 2.0mm holes on shoulders of tomato fruit
- 0.02 mL conidial suspension of \textit{G. candidum} (10^6 spores/mL) inserted into holes using a pipette
- Incubate at 55°F with 80% relative humidity
Need for New Inoculation Methodology?

- Current screening techniques do not reflect natural fruit infections.
- Wounding methods to infect may change the response to postharvest treatments and other experimental procedures.
Proposed New Infiltration Method

1) Disinfect fruit and stem scar
   - Swab with 70% alcohol

2) Apply surfactant and let sit for 5 minutes
   - Silwet-77

3) Pipette spore suspension (10^6 spores/mL) onto stem scar
Proposed New Infiltration Method

4) Put in vacuum chamber
5) Place -0.01 MPa pressure on fruit for 2 minutes
6) Release vacuum for 1 minute, repeat 2x more
7) Incubate at 55°F with 80% relative humidity
New Infiltration Method (cont.)

- Spore suspension infiltrates fruit due to unequal pressures (inside vs. outside)
- There is enough pressure to be pulled into fruit below the stem scar
- Inner fruit → Ideal environment for pathogen growth:
  - Moist
  - Dark
  - Nutrients available
Experiment 1: Testing the Utility of Surfactant

- Stem scars are dry and have high surface tension, which can inhibit the entrance of liquids.
- Does adding a surfactant before inoculation increase the infection rate with the vacuum inoculation method?

- Tomatoes swabbed with surfactant or left dry before applying spore suspension.
- Vacuum inoculation methods followed as stated previously.
Experiment 2: Comparing Wounding and Vacuum Methodology

- Tomato fruit inoculated by either wound or vacuum methods.
- Compare total number of infected fruit and time for symptoms to develop.
Experiment 3: Vacuum method on fruit developmental stages

- There are difficulties infecting non-red tomatoes for research purposes.
- Will vacuum inoculation be more successful at infecting fruit with sour rot?

- Red, breaker (green turning orange), and green tomatoes inoculated using vacuum procedure.
- Rate of infection and total number of fruit infected compared to data from previous studies.
Results: Experiment 1

Resulting infected fruit from vacuum pressure inoculation with and without surfactant (breaker stage fruit).
Results: Experiment 2

Resulting infected fruit from wound and vacuum inoculation (breaker stage fruit).
Results: Experiment 3

Resulting infected fruit from vacuum and wounding inoculation on 3 growth stages of tomato fruit.

*data from wound inoculation of different stages of tomato development from previous study, Steve Rideout, Virginia Tech.
Preliminary Conclusions

- Vacuum inoculation better simulates the physiology of in-field sour rot infections.
- Using surfactant can reduce surface tension from old stem scars to increase infection rates.
- Vacuum inoculation has a higher incidence of infection than wound inoculated fruit.
- Vacuum inoculation results in more infection in green, breaker, and red stages of fruit development than wound inoculation.
Goals for Future Studies

- Investigate efficacy of postharvest chemicals used against sour rot infections using vacuum inoculation versus wounding.
- Use vacuum inoculation to determine susceptibility differences among varieties to *G. candidum*. 
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