P. taeda saplings response to *L. terebrantis* differential inoculum density

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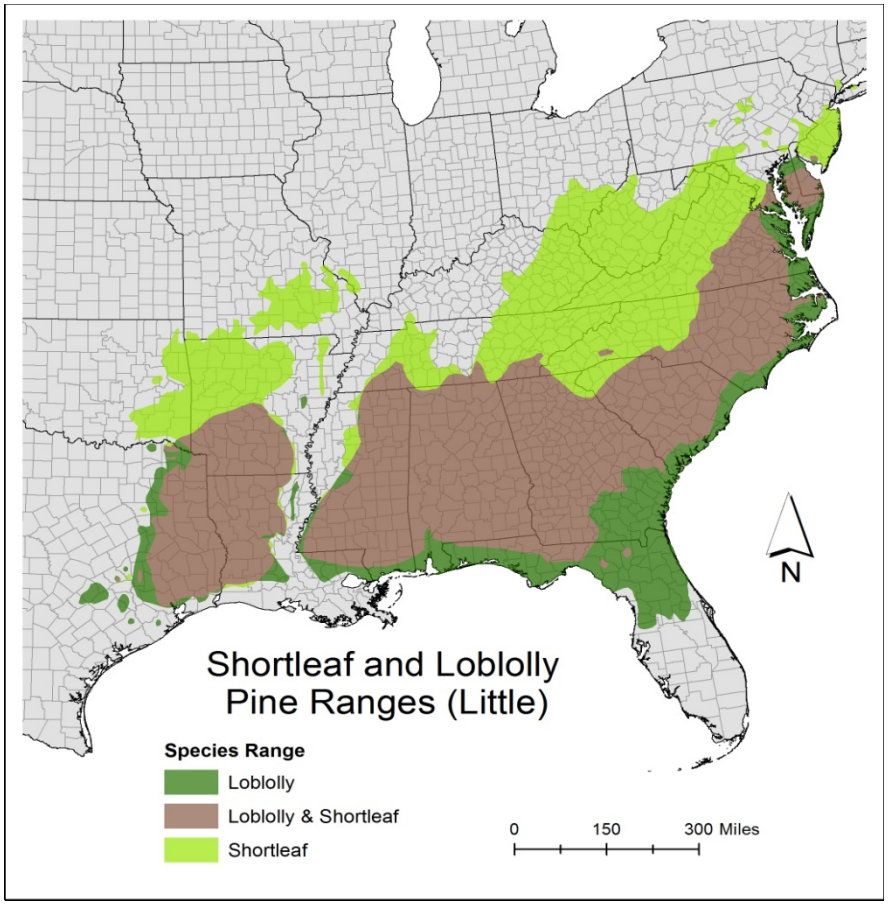
Introduction – *Pinus taeda* (Loblolly Pine)

Veit, J. 2016



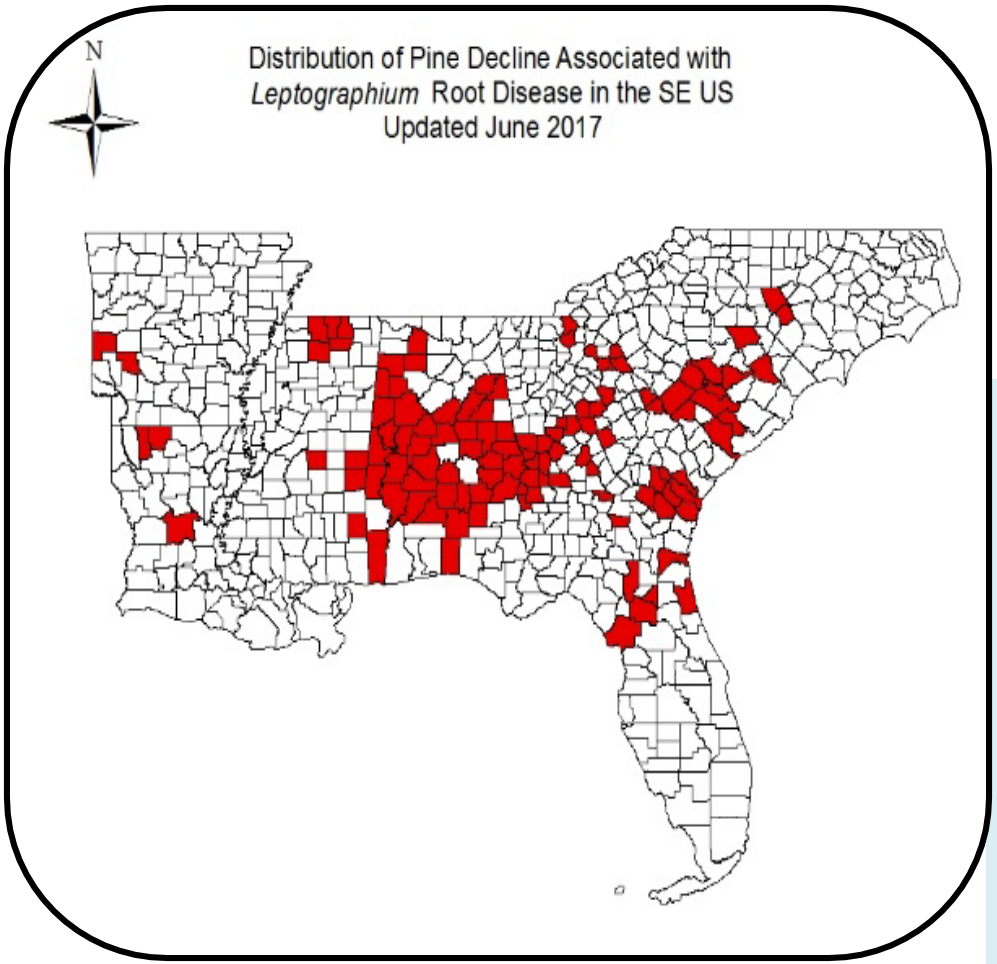
https://en.wikipedia.org/wiki/Pinus_taeda

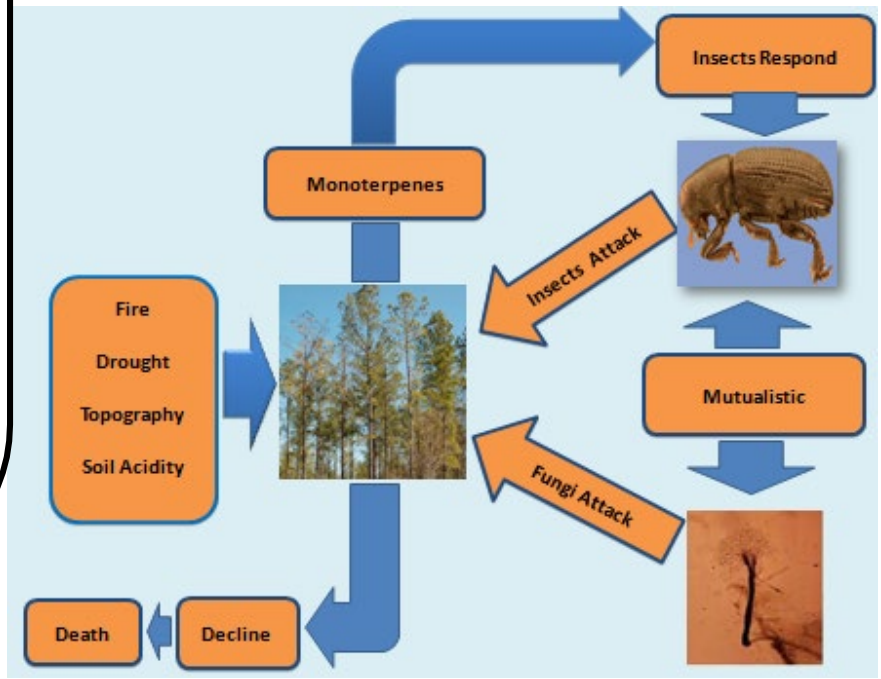
Steven Weaver, SREF

Introduction - Factors Affecting Loblolly Pine

- Abiotic
  - Air pollution
  - Soil erosion
  - Drought/Moisture stress
  - Wind and ice storms
  - Fires
- Biotic
  - Insects pests
  - Fungal pathogens

Introduction - Loblolly Pine Decline





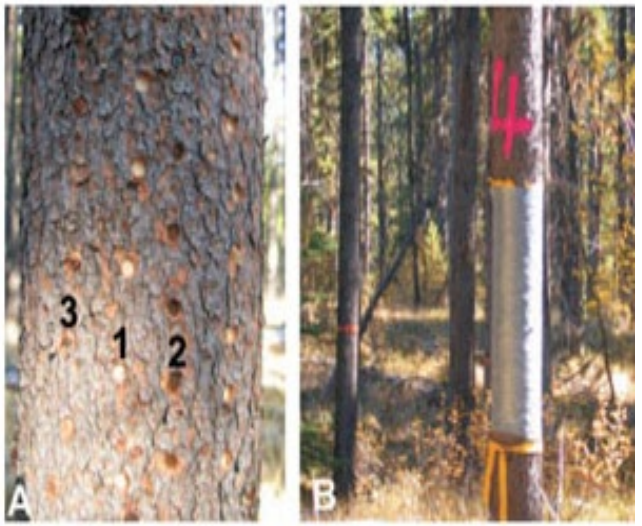
Eckhardt,2003

Questions?

- Does bark beetle associated *Leptographium terebrantis* contribute to loblolly pine decline?
- At what level of the fungal inoculum density does loblolly pine decline occur?
- What are the underlying physiological mechanisms of *L. terebrantis* in loblolly pine decline?
- To what extent does the pathogen alter the host species defensive chemical compounds?

Part 1: Introduction

- Host – pathogen interactions
  - Various inoculation methods
  - Cork borer technique
  - Sterile razor blade



Lee et al, 2006

- Mycelial plugs of 5–12 mm

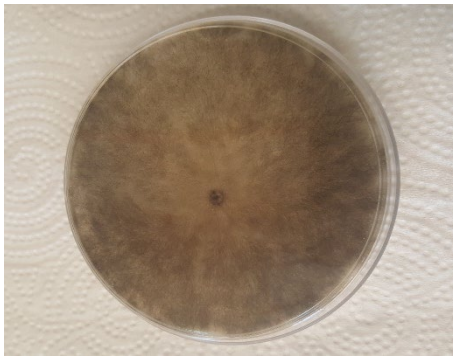
- Introduce large quantity of inoculum

- Vector 2–4 mm long and 1–1.5 mm wide

Introduction

- Assess the suitability of sterile toothpicks (1–2 mm) for the production and transfer of fungal propagules

- Identify inoculum densities for further studies in mature loblolly pine trees

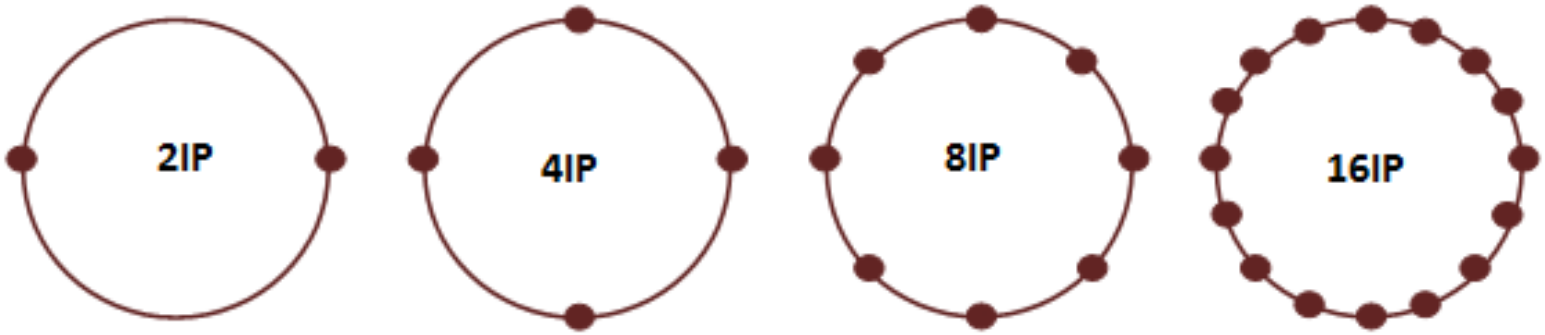


Pure culture of *L. terebrantis*

Conidiophore bearing conidia

Methods

- Study site – Solon Dixon Forestry Center, Andalusia, AL
- Loblolly pine trees 6.4 cm diameters at ground level
- Four fungal treatments
  - Two inoculation points (2IP) at 180^{O}
  - Four at 90^{o} (4IP)
  - Eight at 45^{o} (8IP)
  - Sixteen at 22.5^{o} (16IP)



Levels of fungal treatments

Methods

- Each treatment was randomly assigned to 6 trees with 3 replicates
- Respective control treatment was assigned to 3 trees
- *L. terebrantis* was cultured on sterile toothpicks for 24 days at 23^{o}C
- Post-treatment assessments - 8wks

Inoculation process

Methods

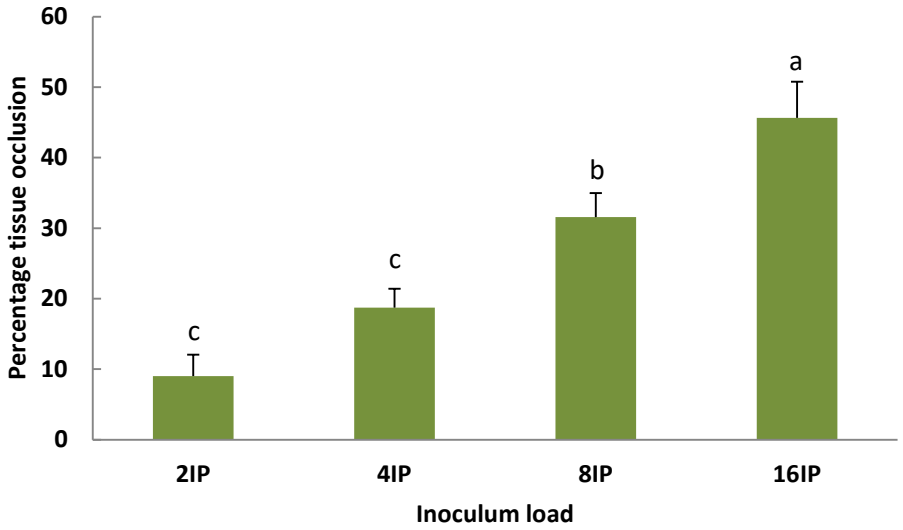
- Trees were cut at ground level and 15 cm above the inoculation zone after 8wks

- Area of tissue occlusion, length and volume were assessed

Cross-sections of trees inoculated with sterile toothpicks

Results

Occlusions produced by *L. terebrantis*



Treatment	Sample size	Occlusion length ± SE (mm)	Occlusion volume ± SE (cm^3)
2IP	72	87.30 ± 5.03a	12.47 ± 1.31a
4IP	72	112.63 ± 7.87ab	29.91 ± 3.19b
8IP	72	116.44 ± 6.64b	52.20 ± 5.11c
16IP	72	144.36 ± 7.88c	106.20 ± 10.43d

Conclusions

- *L. terebrantis* colonized toothpicks served as a useful substrate for the production and transfer of fungal propagules
- Fungal occlusions increased with increasing inoculum density
- Sterilized toothpicks produced negligible tissue occlusions
- The 2IP, 8IP and 16IP were selected for further studies in mature loblolly pine trees

Part 2: Physiological response of *P. taeda* trees to stem inoculation with *L. terebrantis*

- Root pathogen invades host and induces secondary metabolites production
- The metabolites and fungal mycelia cause tissues occlusion and reduce hydraulic function
- Loss of hydraulic function affects physiological process

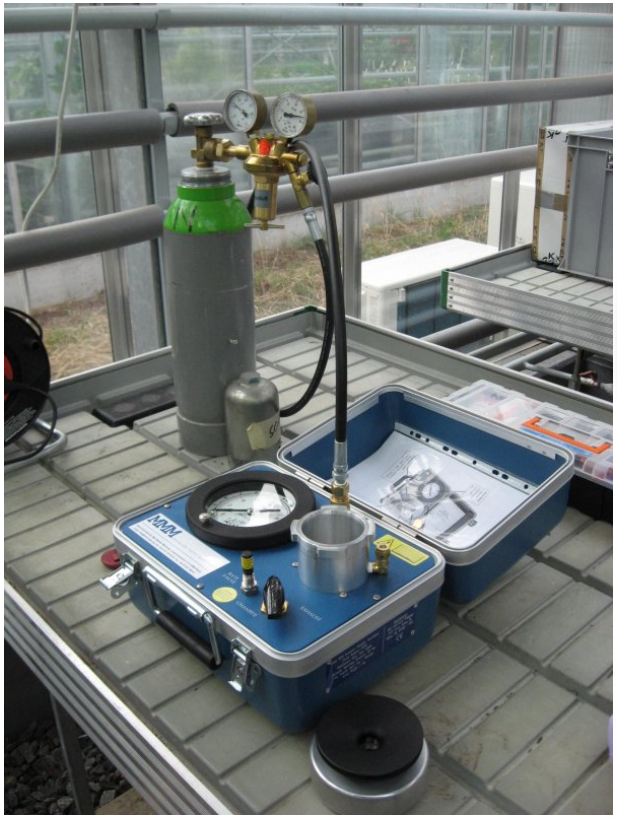
Introduction

- Determine the relationships between pathogen inoculation density, sapwood occlusion and function characterized by hydraulic conductivity and moisture content

- We hypothesized that a positive relationship would be found between *L. terebrantis* inoculation density, sapwood occlusion and the loss of sapwood function

- Pathogen-induced loss of sapwood hydraulic conductivity would cause more negative fascicle predawn water potential and a decrease in stomatal conductance

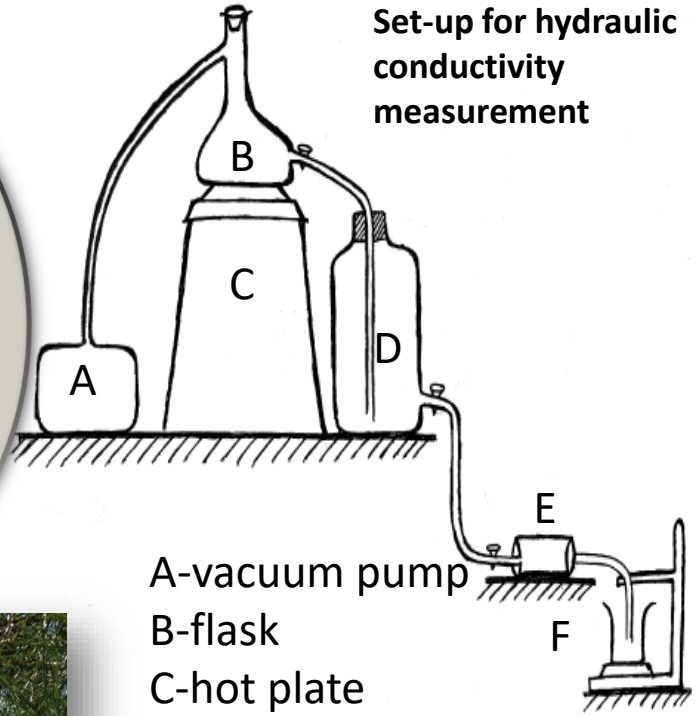
Methods



Pressure Chamber

Porometer

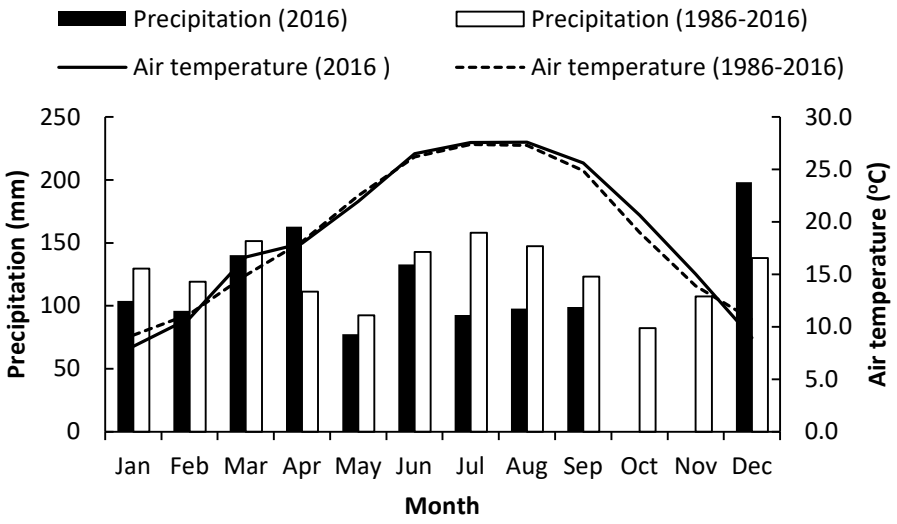
Set-up for hydraulic conductivity measurement



A-vacuum pump
B-flask
C-hot plate
D-water tank
E-stem segment
F-balance and beaker

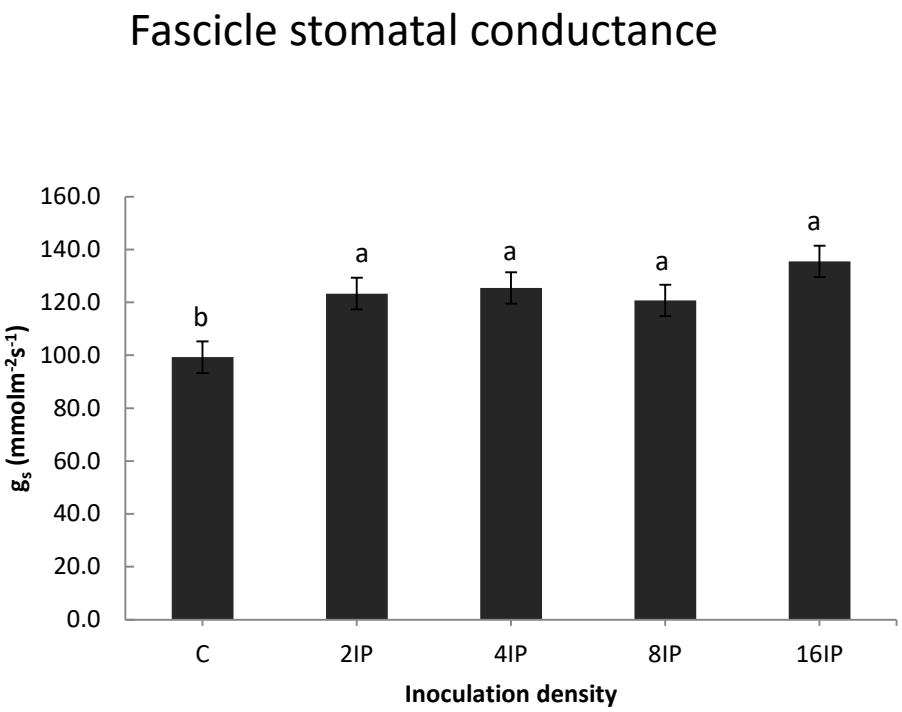
FHDL

Results

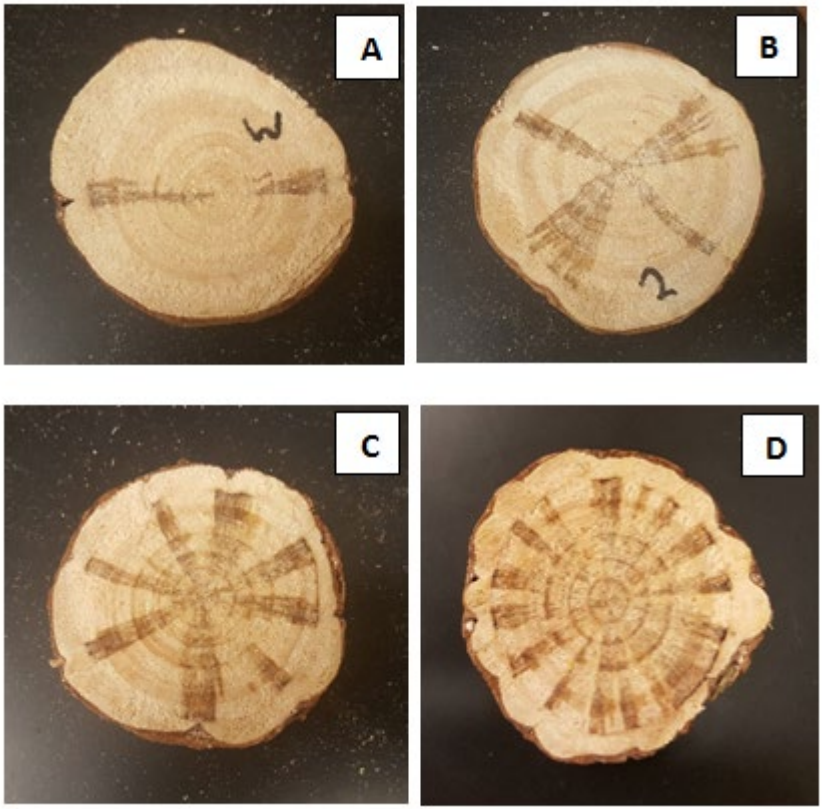


Precipitation and temperature at the site

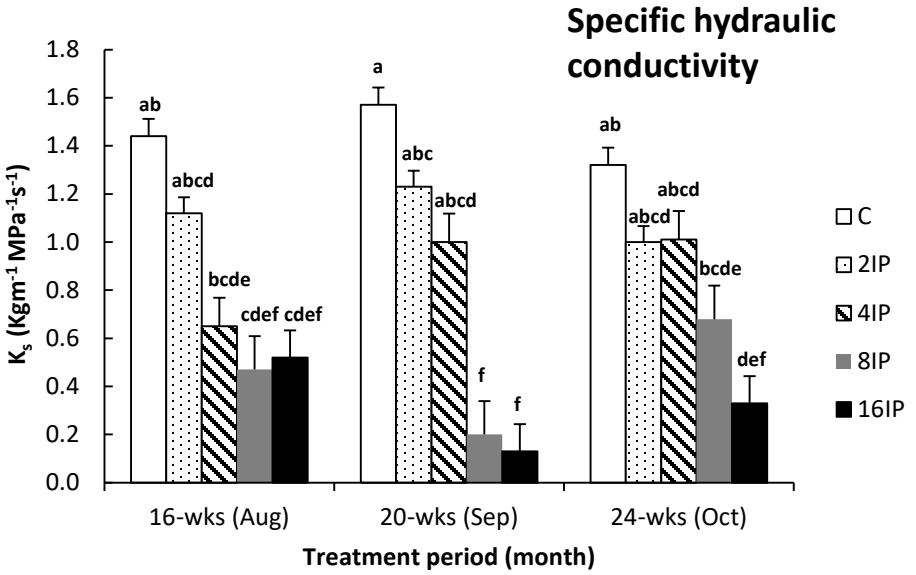
PWP values - ranged between
-0.31 to -0.51 MPa



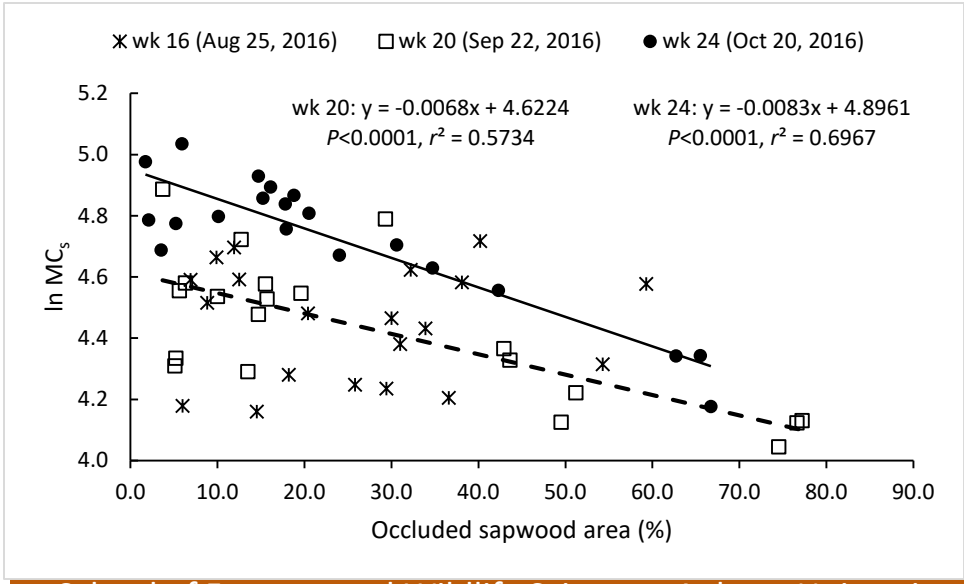
Results



Occlusions of *L. terebrantis*



Moisture content and occlusion area relationship



Conclusions

- The young *P. taeda* trees tolerated *L. terebrantis* infection by producing producing new sapwood
- In support of our first hypothesis, we observed decreases in stem hydraulic conductivity and moisture content as occluded sapwood area increased
- We reject the second hypothesis that pathogen induced loss of hydraulic conductivity will decrease stomatal conductance
- Elevated stomatal conductance during the study is indicative of high rates of carbon fixation
- Relationships between occluded sapwood area and both stem hydraulic conductivity and moisture content suggests that there are underlying risks to *L. terebrantis* infection

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