



# Evaluating biological control for *Sclerotinia* stem rot of canola



Megan McCaghey, Assistant Professor

University of Minnesota, Twin Cities

Coauthors: Jasper Tao, Donn Vellekson, Dave  
Grafstrom, Peter Aspholm, Carah Anteck,  
Nancy Ehlke,

2024 Canola Symposium

# Sclerotinia stem rot

## *Sclerotinia sclerotiorum*



[www.canolacouncil.org](http://www.canolacouncil.org)

### Sclerotinia Stem Rot Disease Cycle

(Caused by the fungus *Sclerotinia sclerotiorum*)

#### 3 Ascospore Distributes on Petals

The windborne ascospores adhere to flower petals and other organic material.

#### 4 Germination and Distribution of Infection

Ascospores germinate, infect the petal, and spread to adjacent tissues of healthy leaves and stems by direct contact.

#### 5 Distribution of Fungal Lesion

The lesions progress up and down the stem. At this stage, wilted leaves can be visible.

#### 6 Formation of New Sclerotia

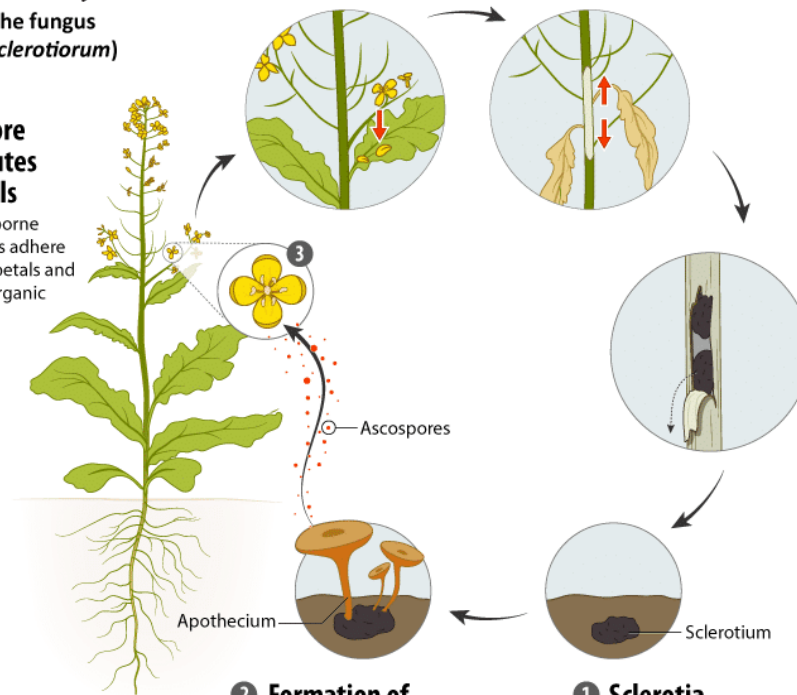
The infected stem becomes bleached and brittle and forms new sclerotia. The sclerotia return to the soil at harvest and the cycle repeats.

#### 2 Formation of Apothecia

Spore-producing apothecia germinate from sclerotia under moist plant canopy and release ascospores.

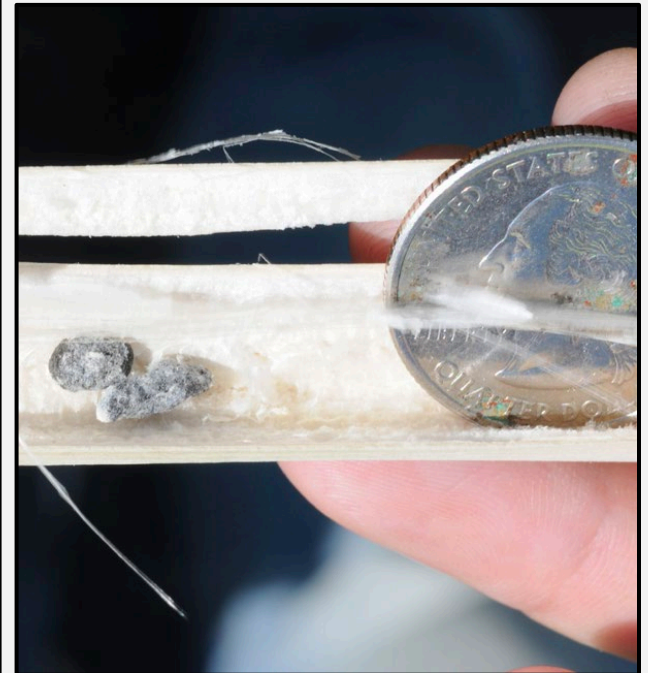
#### 1 Sclerotia Overwinter in Soil

The stem rot fungus (*Sclerotinia sclerotiorum*) overwinters as sclerotia in the soil or in stubble at the soil surface.



# Sclerotinia stem rot

## Signs and symptoms in canola



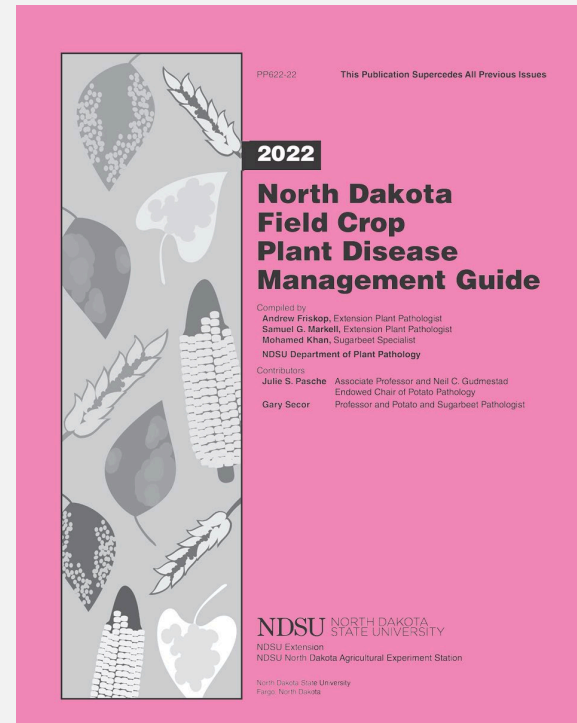
# Sclerotinia stem rot in canola- management

- Fungicide application timing is critical (20-50% of flowering)- look for apothecia
- Forecasting tools are available
- Various options: Ex. Boscalid and Azoxystrobin AI
- Hybrids with tolerance and upright architecture
- Rotation with non-susceptible crops (grasses) to reduce inoculum (does not eliminate)

Dr. Sam Markell

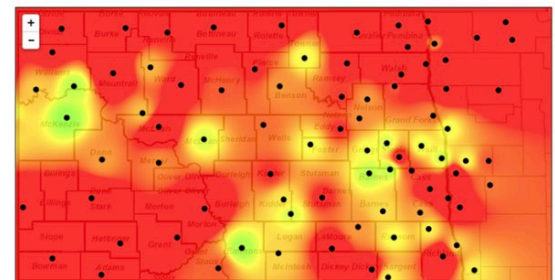
Dr. Luis Del Rio

[www.ag.ndsu.edu](http://www.ag.ndsu.edu)



## Sclerotinia Forecast Map

Estimated risk of Sclerotinia stem rot development for 9/7/2022



# Translational tools towards SSR management

1

Integrated management of soilborne diseases- pathogen ecology considerations

2

Enhancing white mold/*Sclerotinia* tolerance through genetic and architectural strategies

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# Biological control products to manage *Sclerotinia* stem rot in canola

Biofungicide mechanisms to control pathogens:

- 1) The production of toxins (antibiosis)
- 2) Parasitism that attacks the pathogen
- 3) Competition for resources
- 4) Induced resistance of the crop plant

## **Benefits:**

- Can be used in organic production (premium price)
- Can be used with conventional fungicides to avoid resistance
- Some may have long lasting effects
- Short reentry period

## **Challenges:**

Application timing is key and results may vary

# Biological control products to manage Sclerotinia stem rot in canola



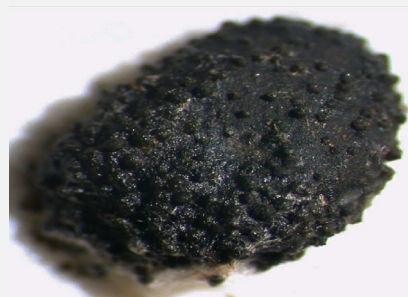


# Biological control products to manage *Sclerotinia* stem rot in canola



## *Coniothyrium minitans*

- A mycoparasite of sclerotia (reduction by as much as 95%), will persist in the soil
- Applied to the soil — pre-plant, at planting, after cultivation or post-harvest
- Can be applied at higher rate in spring (3-4 lb) or lower rate in fall (1-2 lb): must contact sclerotia



# Biological control products to manage *Sclerotinia* stem rot in canola



## *Bacillus subtilis*

- Applied during flowering
- Works as an antagonist (antibiosis)
- Stops the growth of mycelia and germination of ascospores
- May activate host defense response

# Biological control products to manage *Sclerotinia* stem rot in canola

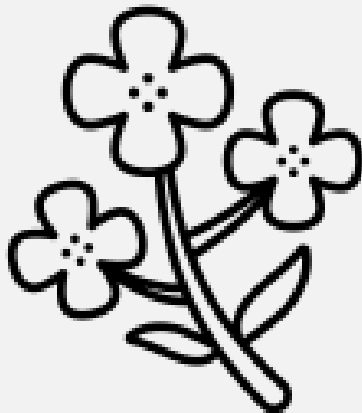


## *Bacillus amyloliquefaciens*

- Applied during flowering or to soil
- Antibiosis: Inhibits mycelial growth, suppresses of sclerotia formation, reduces ascospore germination, and induces structural abnormalities within the apothecia
- Plant growth-promotional activity
- Suppression comparable to conventional fungicides in snap bean (Pethybridge et al., 2019)

# Objectives to evaluate biological control products to manage *Sclerotinia* stem rot in canola

- 1) Compare the efficacy of current, diverse biological control products at reducing disease and inoculum
- 2) Compare the impact of biological control products on yield
- 3) Evaluate the potential of fungi and bacteria isolated from sclerotia for novel biological disease control



# Obj. 1: Compare the efficacy of current biological control products



Carah Anteck



- Trials were conducted at UMN St. Paul and Roseau CPC farms (planted 4/23 and 5/29)
- Each treatment was replicated four times in an RCBD
- Plots were 25' long and 6' wide
- Irrigation was applied through early flowering in SP (June 7)
- Sclerotia were spread to equal about 1 per sq ft

# Biological control agents, rates, and timings

Treatment	Product	Active Ingredient	Application Timing	Application Rate
1	Untreated control	NA	NA	NA
2	Contans	<i>C. minitans</i>	May, pre plant, soil application	4 lb/ac
3	Double Nickel LC	<i>B. amyloliquefaciens</i>	20-30% bloom	5 qt/ac
4	Double Nickel LC	<i>B. amyloliquefaciens</i>	50-60% bloom	5 qt/ac
5	Double Nickel LC	<i>B. amyloliquefaciens</i>	20-30% bloom + 7 days later	5 qt/ac
6	Serenade OPTI	<i>B. subtilis</i>	20-30% bloom	20 oz/ac
7	Serenade OPTI	<i>B. subtilis</i>	50-60% bloom	20 oz/ac
8	Serenade OPTI	<i>B. subtilis</i>	20-30% bloom + 7 days later	20 oz/ac
9	Contans + Serenade OPTI	<i>C. minitans</i> + <i>B. subtilis</i>	May (pre plant soil app) + 20-30% bloom	4 lb/ac + 20 oz/ac
10	Endura*	Boscalid	20-30% bloom	5.5 oz/ac
11	Proline, Endura*	Prothioconazole, boscalid	20-30% bloom + 7 days later w/ Endura	5.5,5.7 oz/ac

# Disease evaluation methods

- Disease was assessed from 15' of each row at **full pod** (July 3<sup>rd</sup>, St. Paul and August 6<sup>th</sup>, Roseau)
- Both incidence and severity data were collected

$$\text{DSI} = 100 * [ (\# \text{plants} * 1) + (\# \text{plants} * 2) + (\# \text{plants} * 3) + (\# \text{plants} * 4) + (\# \text{plants} * 5) ] / (\text{Total number of plants in the sample} * 5)$$



1

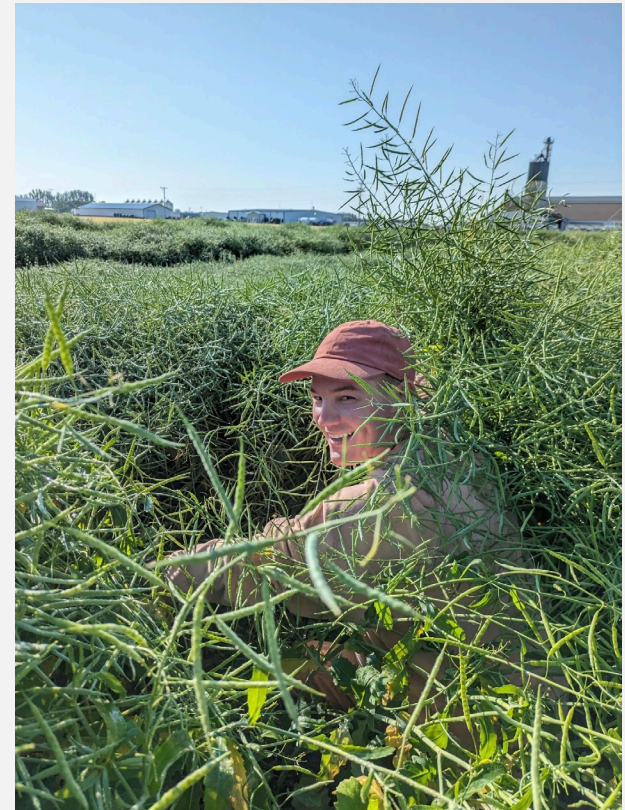
2

3

4

5

[Sclerotiniariskcalculator.com](http://Sclerotiniariskcalculator.com)

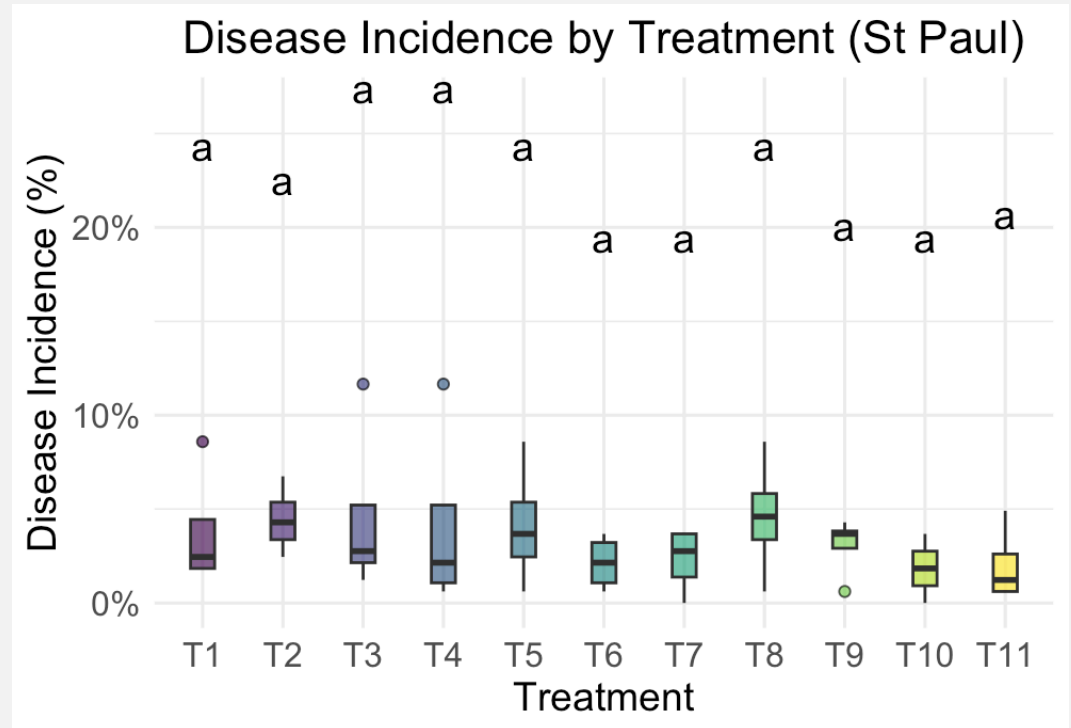


# Disease incidence- St. Paul



Jasper Tao

Trt	Product	Application Timing
1	Untreated control	NA
2	Contans	May, pre plant soil application
3	Double Nickel LC	20-30% bloom
4	Double Nickel LC	50-60% bloom
5	Double Nickel LC	20-30% bloom + 7 days later
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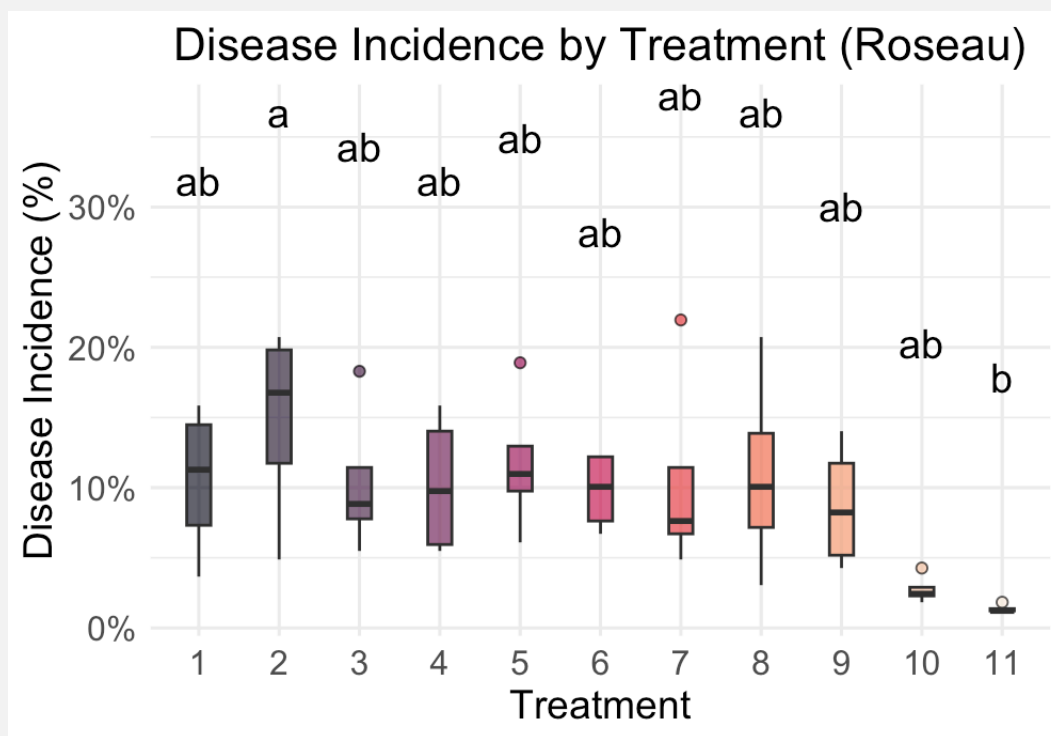


Disease incidence did not differ between treatments.



# Disease incidence- Roseau

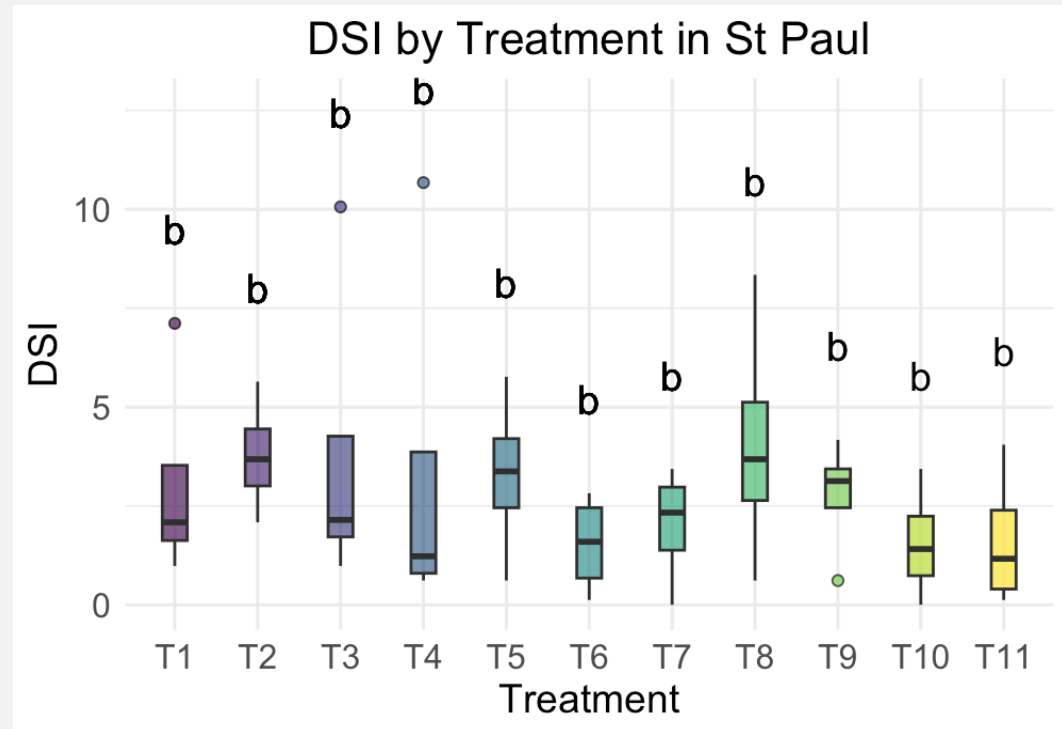
Trt	Product	Application Timing
1	Untreated control	NA
2	Contans	May (pre plant soil application)
3	Double Nickel LC	20-30% bloom
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Disease incidence was lowest in plots treated with premium fungicides (compared to Contans)

# Disease severity index – St. Paul

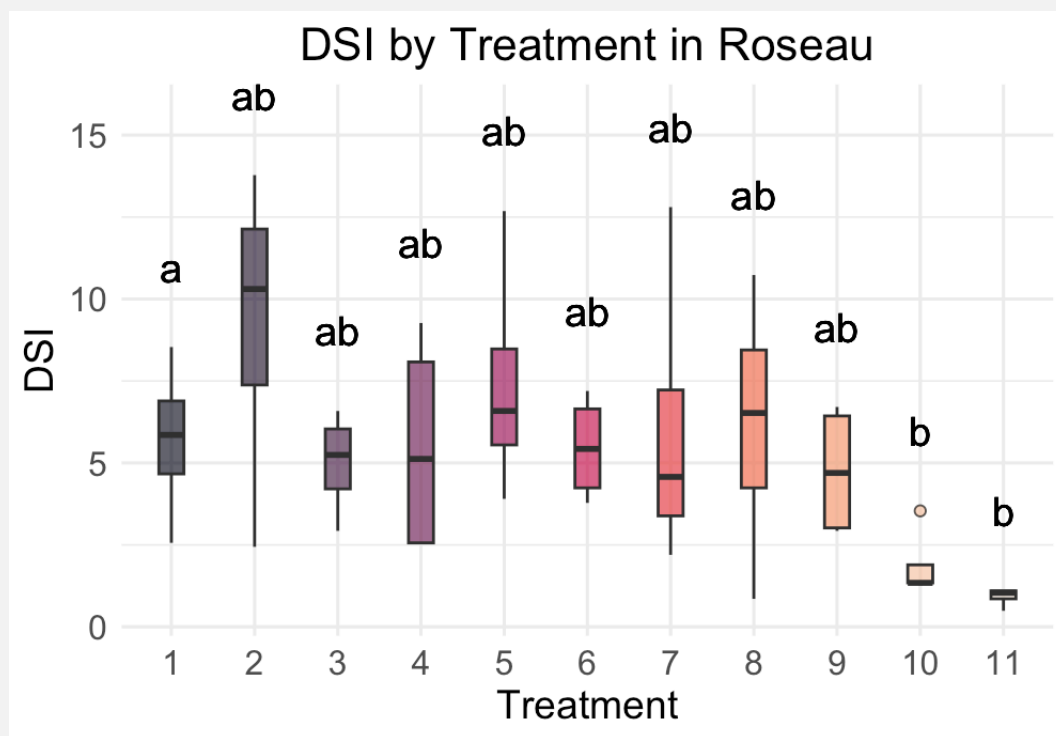
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DSI did not differ between treatments.

# Disease severity index – Roseau

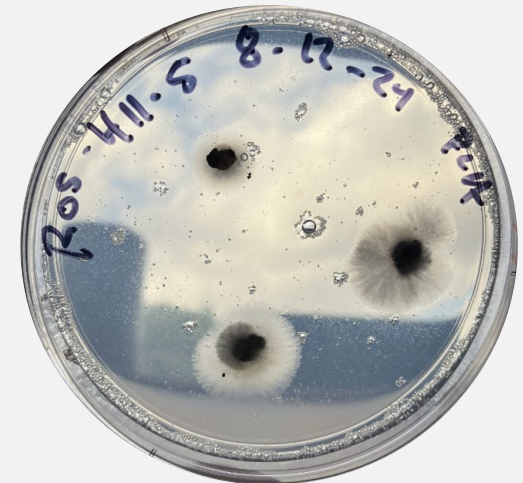
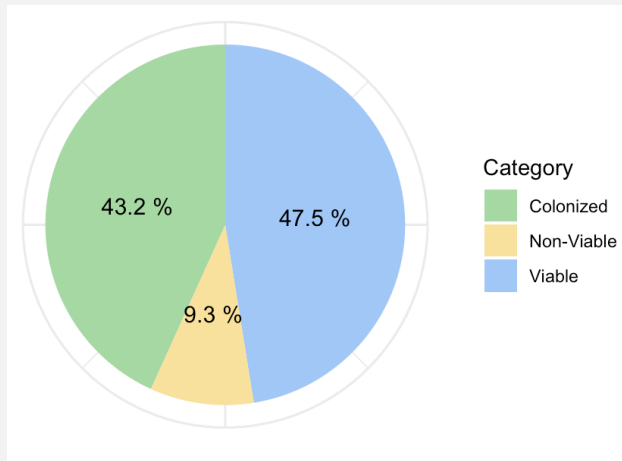
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DSI was lowest in plots treated with Endura and premium fungicides compared to the control

# Inoculum persistence

- At planting, bags of sclerotia (15 sclerotia per bag) were buried 2.5" in the untreated control and Contans- only containing plots
- Bags were removed at harvest



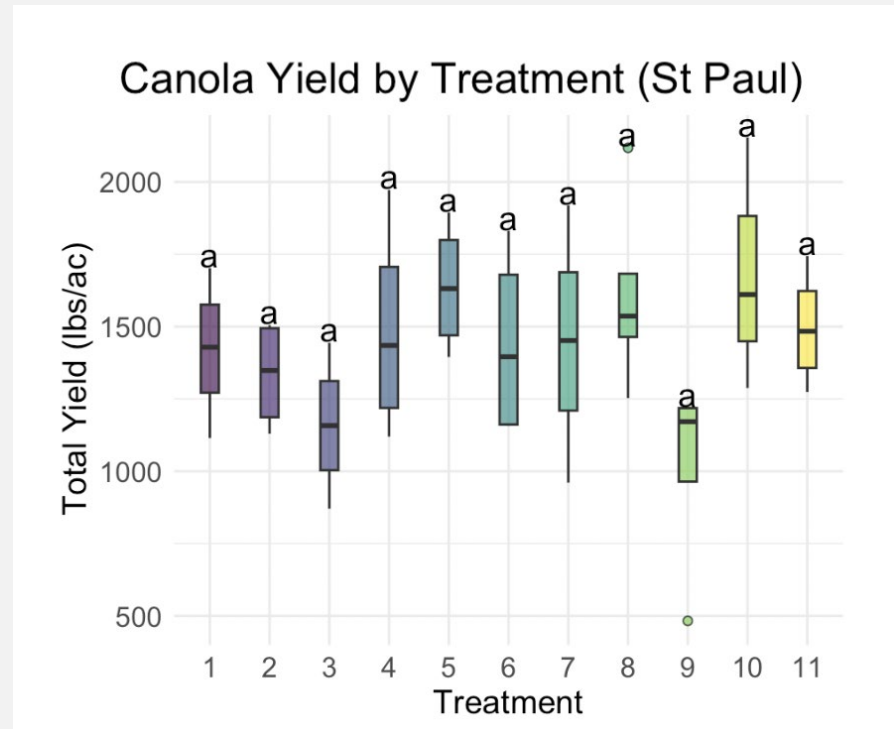


**Obj. 2:  
Compare the  
impact of  
biological  
control products  
on yield**

- Plots were harvested (Aug. 14<sup>th</sup>, St. Paul and Sept. 15<sup>th</sup>, Roseau).
- Plots were measured to increase the precision of yield measurements.
- Yield values were converted to 8.5% moisture.
- Grain quality metrics (oil and protein) were also assessed- *no significant difference between treatments*

# Yield- St. Paul

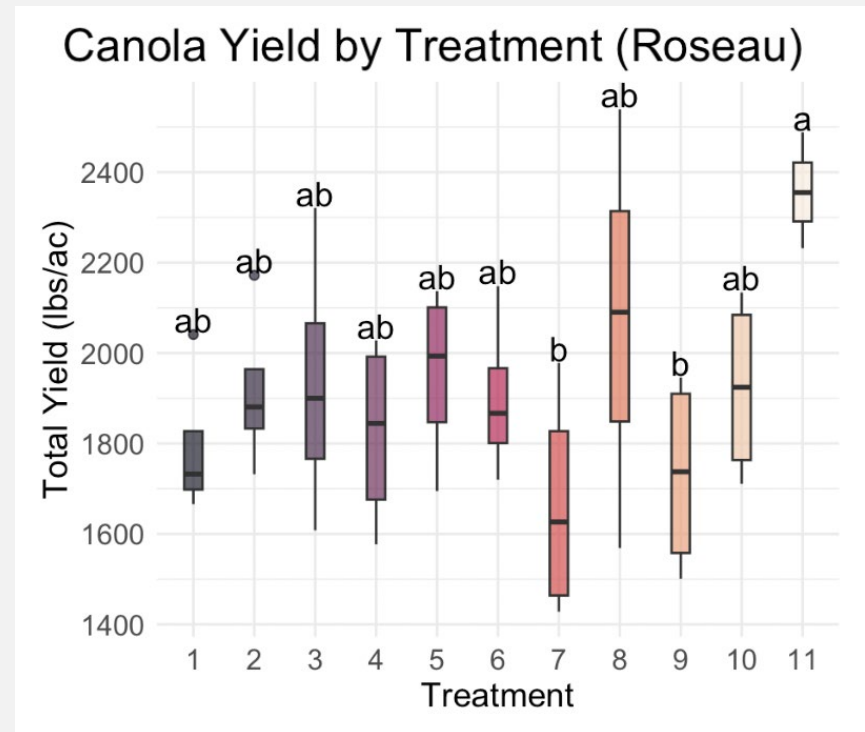
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Yield did not differ between treatments.

# Yield- Roseau

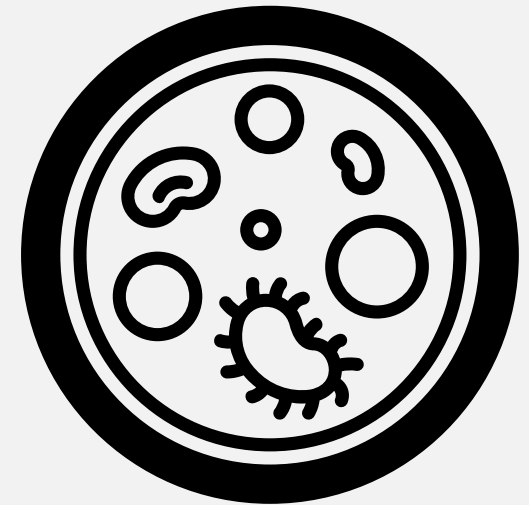
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Premium fungicides resulted in higher yields than two Serenade treatments.

# Obj: 3 Evaluate the potential of fungi and bacteria isolated from sclerotia for novel biological disease control

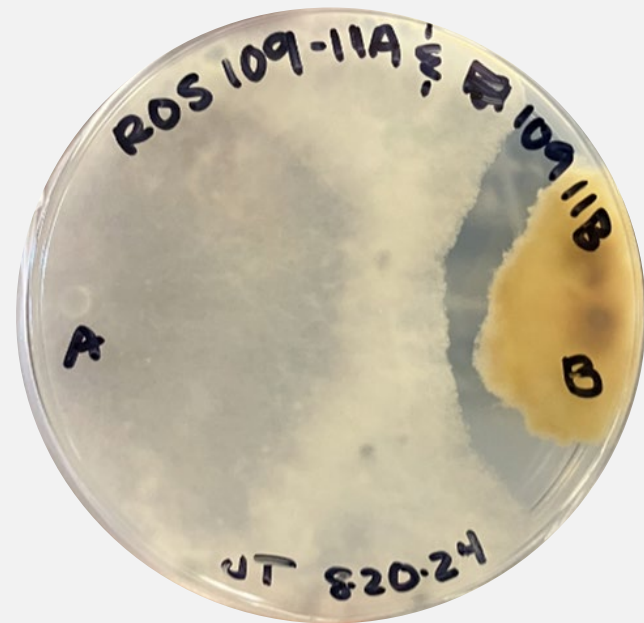
- Buried sclerotia were removed at harvest
- 18 sclerotia per plot were plated directly on fungi culture medium (9) or washed for bacteria (9)
- Pure cultures will then be grown in inhibition assays with *S. sclerotiorum* to observe antagonistic activity
- Fungi and bacteria with observed antagonism will be identified using molecular methods: DNA extraction and sequencing with ITS 1 and 4 primers (for fungi) and 16s (for bacteria)





# Microbial isolations

- Inhibition assays will be conducted this winter: 68 unique bacterial isolates, 12 unique fungal isolates
- 12 isolates showed inhibition from viability assays!



# Realized and expected outcomes:

- Biological control products (when evaluated at an incidence less than 20%) are not more effective at disease suppression than conventional fungicides
- We are on the path to identifying novel antagonists from disease inoculum

## NEXT STEPS

- Evaluations with sprayed inoculum?
- Biofungicides + conventional fungicides?
- Antagonism assays with co-plating and buried sclerotia bags in controlled conditions

# Translational tools towards SSR management

1

Integrated management of soilborne diseases- pathogen ecology considerations

2

Enhancing white mold/*Sclerotinia* tolerance through genetic and architectural strategies

# Evaluating Ss aggressiveness determinants across crop species

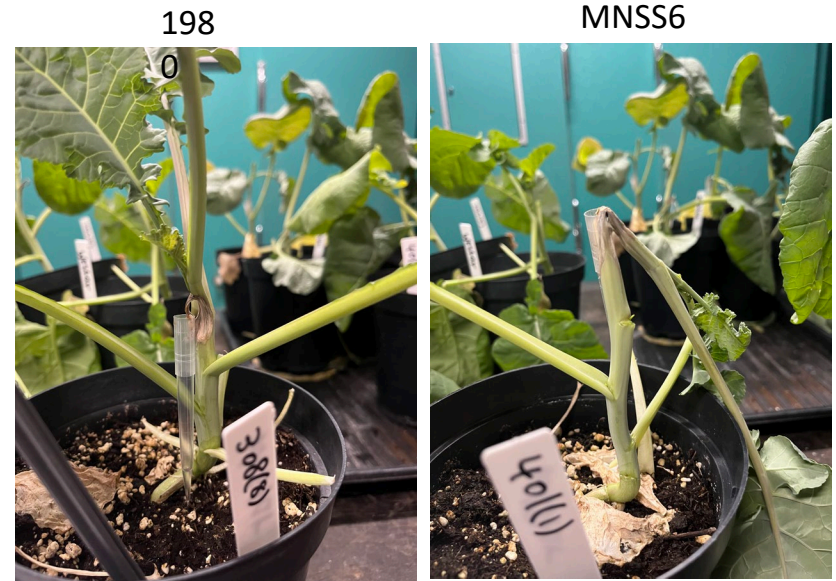


Hsuan Fu Wang



# Development of a screening panel for multiple crop species

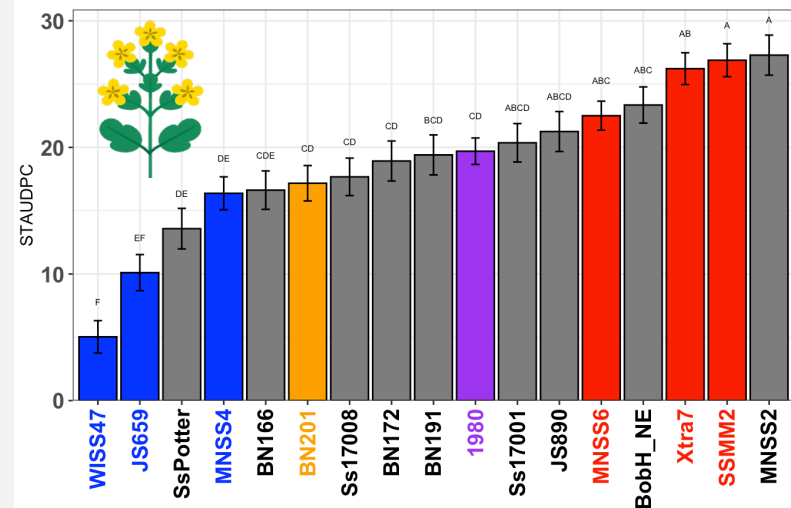
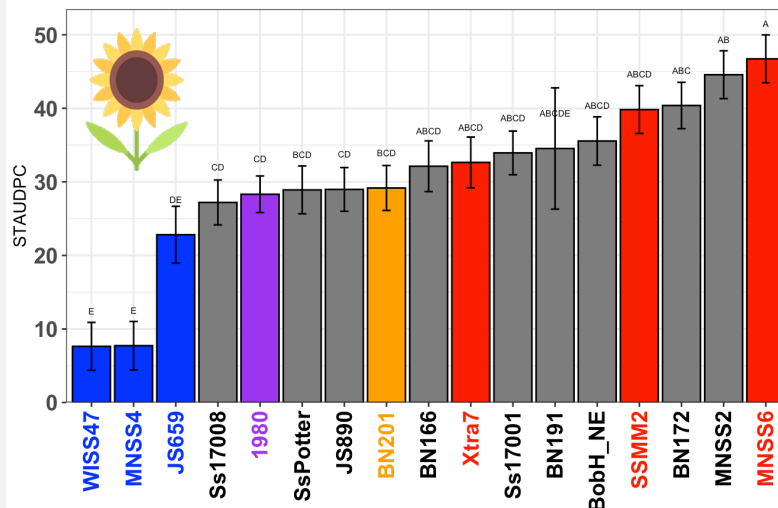
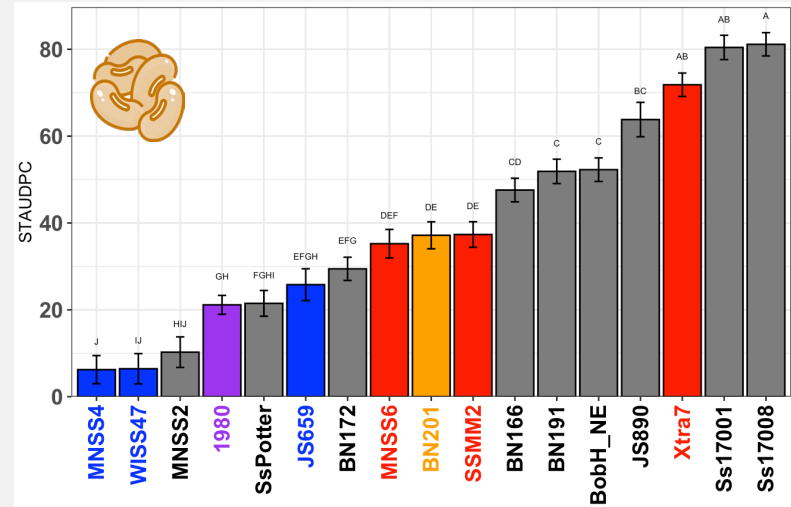
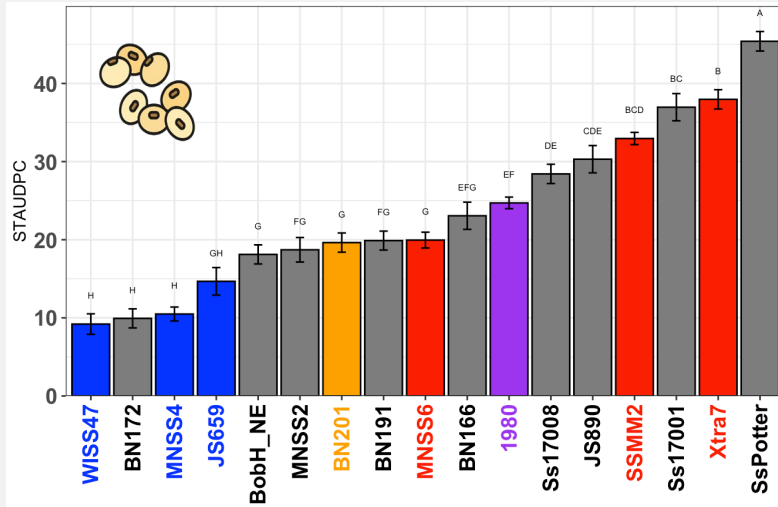
- Isolates of *S. sclerotiorum* differ in their aggressiveness
- Each isolate (17 so far) was repeated 6 times on a susceptible variety
- Inoculated at V4-V5
- Third leaf excised at 0.75"



Cultivar	Plant (Scientific name)	Source
PI605719(Flint)	Canola ( <i>Brassica napus</i> )	North Central Regional PI Station (NC7)
PI649150 (Westar)	Canola ( <i>Brassica napus</i> )	North Central Regional PI Station (NC7)



# Development of a screening panel for multiple crop species



# Evaluate *Ss* aggressiveness determinants across crop species- **screening panel and transcriptomics to ID RNAi targets**



Hsuan Fu Wang



# Realized and expected outcomes:

- A *S. sclerotiorum* isolate collection
- Improved screening tools for breeders
- Targets for gene silencing that can have broad impact across crop species

## NEXT STEPS

- Development of a subpanel and screening for resistance in R vs. S lines
- Transcriptomics to identify aggressiveness-related genes!



# THANK YOU

## *Questions?*

### Special thanks to:

Donn Vellekson

Dave Grafstrom

Dr. Nancy Ehlke

Peter Aspholm

Carah Anteck

Jasper Tao

Hunter Kluegel

Alisha Mildenberger



**Minnesota  
Canola Council**



**National  
Sclerotinia  
Initiative**

Program of the



- Additional ideas or thoughts?

*Please email me: [mmccaghe@umn.edu](mailto:mmccaghe@umn.edu)*

# Additional production notes

Variety- InVigor L345PC

Fungicide application – 18 GPA @28 PSI

Seed treatment- Helix Vibrance

Nutrient- PPI 140-40-40-20 R, 100-0-0-20 St Paul