



# ISOLATION AND PATHOGENICITY OF *PHOMOPSIS* FROM SYMPTOMLESS SUNFLOWER

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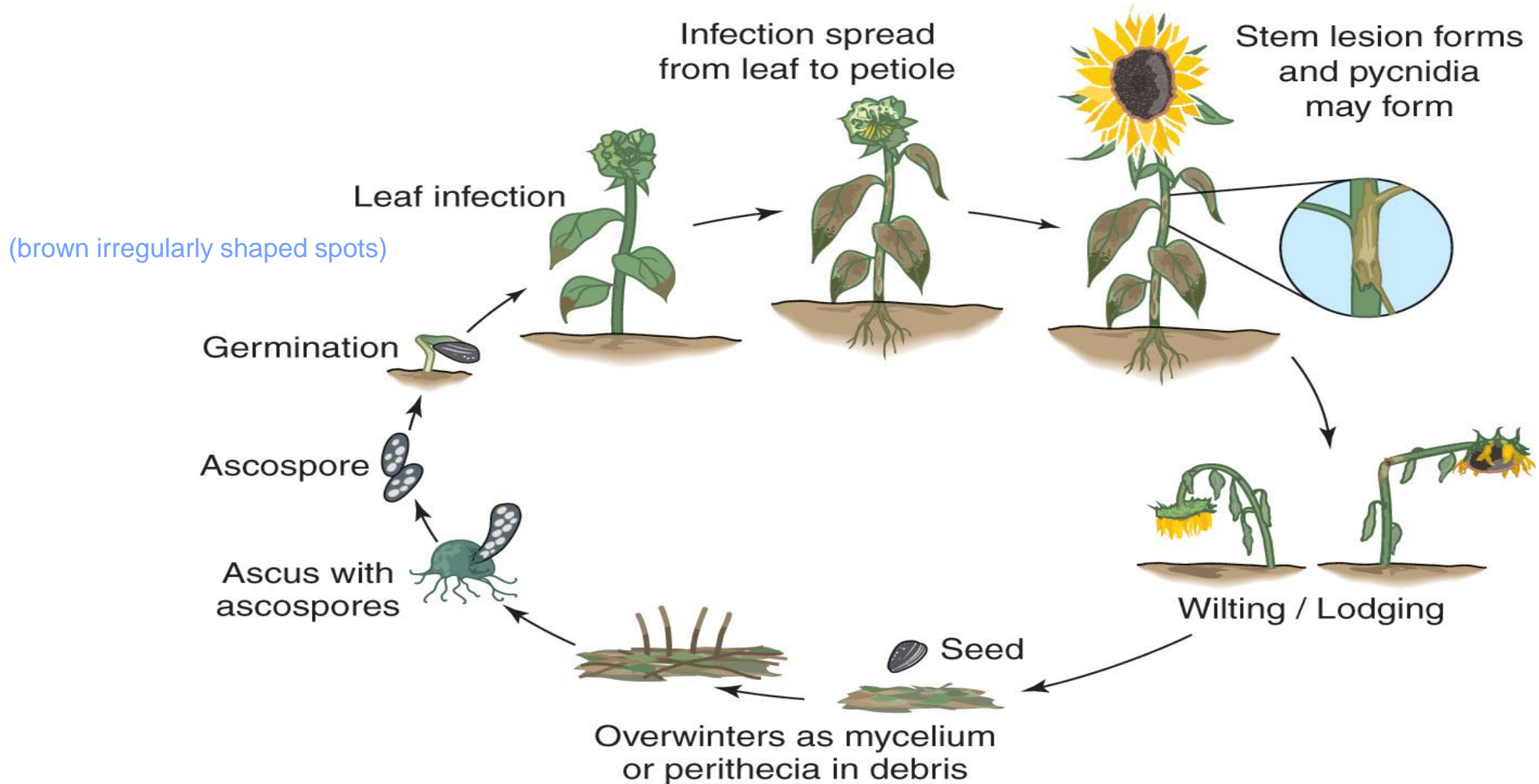


# BACKGROUND

- **Phomopsis stem canker**
- Causal pathogens described in the U.S.:
  - *Diaporthe helianthi* (syn. *Phomopsis helianthi*)
  - *D. gulyae*
  - *D. stewartii*

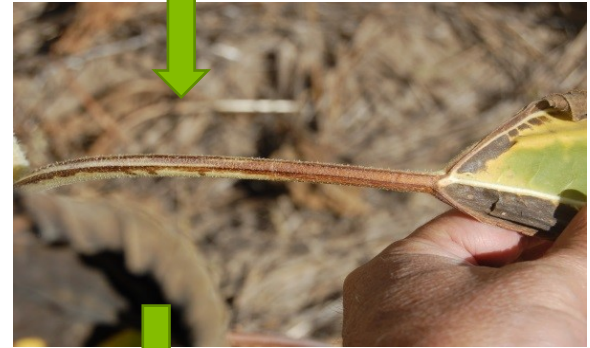


# DISEASE CYCLE



# PREVIOUS RESEARCH

- Fungus enters through leaves, progresses towards the petioles and finally enters the stem.
- Stem lesions originate from internal fungal development.



Source: Vukojević et al. 2001



# JUSTIFICATION

- Studies related to management options (e.g. fungicides) showed inconsistent results
- One of the hypothesis is species of *Diaporthe* may be “latent” as described in other crops, such as soybean (Kmetz et al. 1979).



# RESEARCH OBJECTIVES

- Examine the possibility of latent infection by species of *Diaporthe* on sunflower
- Determine if “latent” species of *Diaporthe* are pathogenic on sunflower



# MATERIALS AND METHODS

## ➤ **Field study:**

- Location: ND, NE and SD (One site each)
- Large plots (50 ft long by 10 ft wide)
- 4 replicate blocks
- Sunflower: Rh 400 CL
- Plant samples taken biweekly basis, cut into parts (stems, leaves, roots) and air-dried.
- Heads sampled during maturity



# MATERIALS AND METHODS

- Fungal isolation and identify confirmation
  - 6 pieces of stem tissue randomly selected
  - Surface-sterilized and placed on PDA
  - Incubated
  - Fungi isolated and purified
  - Identified by morphology and qPCR (Elverson et al. 2019)





# RESULTS – SOUTH DAKOTA

## ➤ Symptom observed in field:

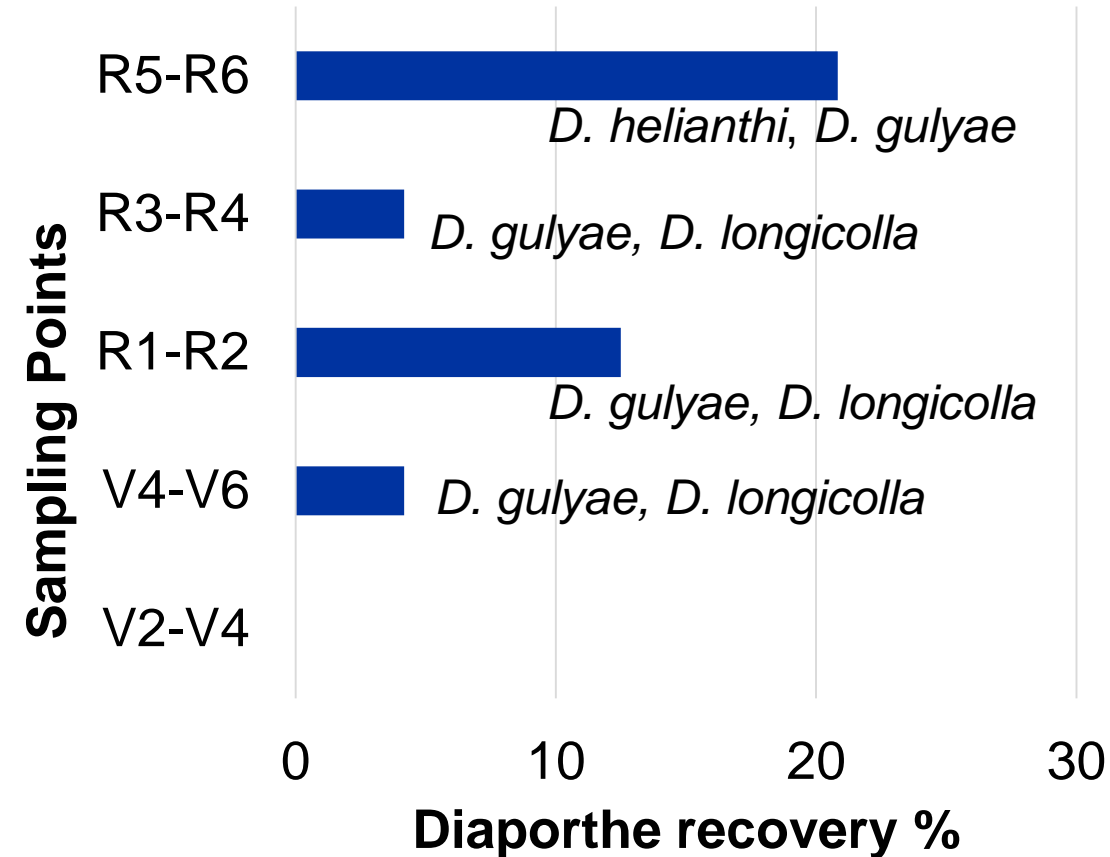
1<sup>st</sup> SP (V2-V4): No disease

2<sup>nd</sup> SP (V4-V6): No disease

3<sup>rd</sup> SP (R1-R2): Few lower leaves had symptoms

4<sup>th</sup> SP (R3-R4): Few petioles had symptoms

5<sup>th</sup> SP (R5-R6): Symptom in lower leaves, petioles and stems



# MATERIALS AND METHODS

## Greenhouse experiment:

- Design: CRD, 6 pots per treatment (2 plants in each pot)
- Treatments: 7 isolates and a non-inoculated control



Treatments	Species	Sampling point
N-58	<i>D. gulyae</i>	3 <sup>rd</sup>
N-31	<i>D. longicolla</i>	4 <sup>th</sup>
N-4	<i>D. gulyae</i>	5 <sup>th</sup>
N-1, N-3, N-5, N-6	<i>D. helianthi</i>	5 <sup>th</sup>
Non-inoculated control	Sterilized PDA plug	

# MATERIALS AND METHODS

Greenhouse experiment.....

- Inoculation at R1 stage (The terminal bud forms a miniature floral head rather than a cluster of leaves)
- Stem wound method (Mathew et al. 2015)
- Inserted a mycelial plug (4 mm)
- Covered with petroleum jelly
- Misting for 3 days





# MATERIALS AND METHODS

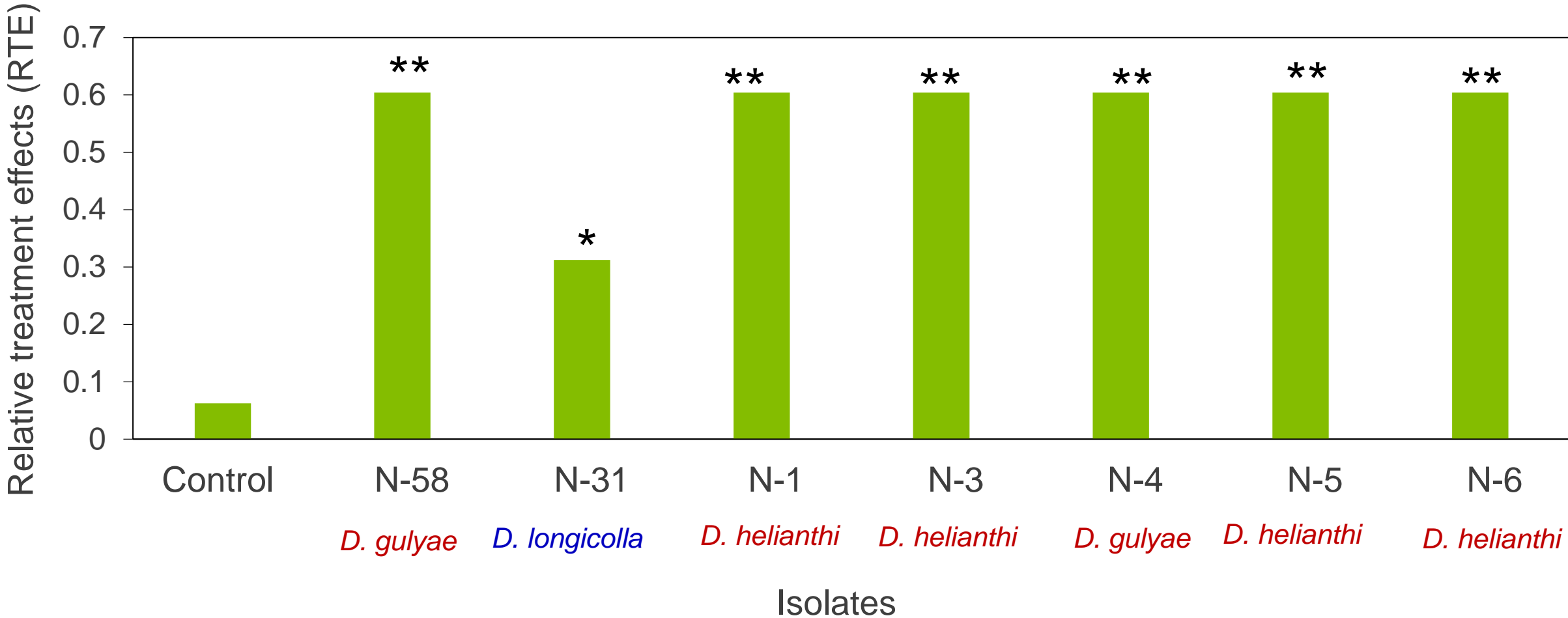
Greenhouse experiment....

- At 15 days after inoculation, Disease severity scored using a **0 to 5** scale (Mathew et al. 2015)
- Data analyses using non-parametric statistics in R.





# RESULTS



# RESULTS



*D. longicolla*



*D. gulyae*



*D. gulyae*



*D. helianthi*

# SUMMARY

- Species of *Diaporthe* cause latent infection of sunflower at early vegetative stages
  - *D. gulyae* and *D. longicolla*
  - First report of *D. longicolla* causing disease
- These pathogens cause disease at R1 and favorable environment



# FUTURE WORK

- Isolates of *Diaporthe* recovered from stem (ND, NE) and roots (ND, NE and SD)
- Identity of the fungus and pathogenicity study in progress
- Repeat the etiology study in 2020 in ND, NE and SD.





THANK YOU



NDSU



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