Molecular epidemiology of Cercospora leaf spot

Detection of latent CLS in commercial sugarbeet fields and monitoring fungicide resistance mutations

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U.S. DEPARTMENT OF AGRICULTURE

Cercospora beticola

- Cercospora leaf spot (CLS) on sugarbeet
- Hemibiotrophic fungus
  - Has a latency period prior to symptoms
- Polycyclic lifecycle
- Genetically diverse
- Primarily controlled through fungicide applications and host resistance

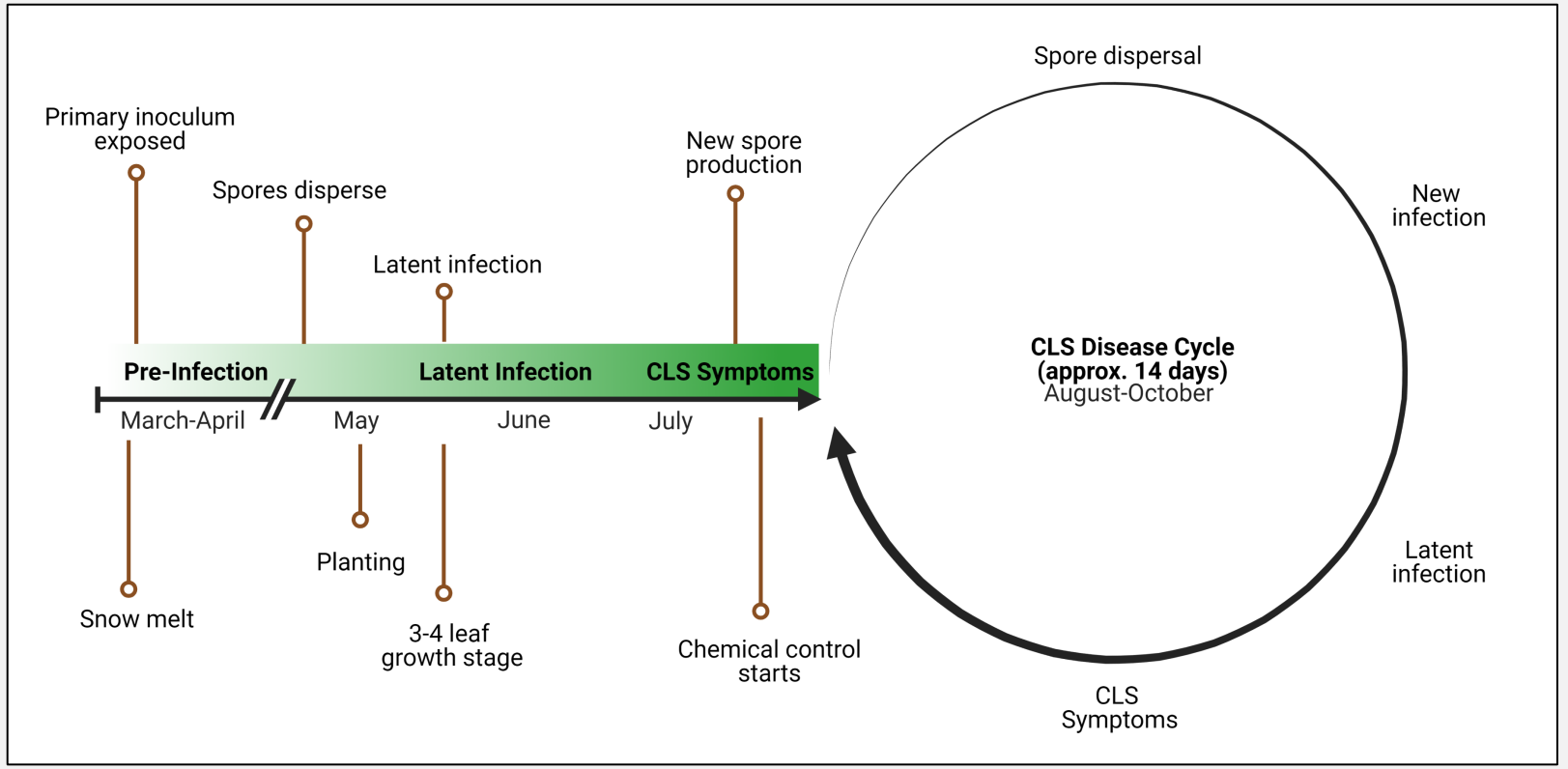
CLS disease cycle

When are spores dispersed in spring?
Detected beginning of May.

What conditions are favorable for spore germination?
Temperatures > 50 degrees F
Available free water (Rain/Dew)

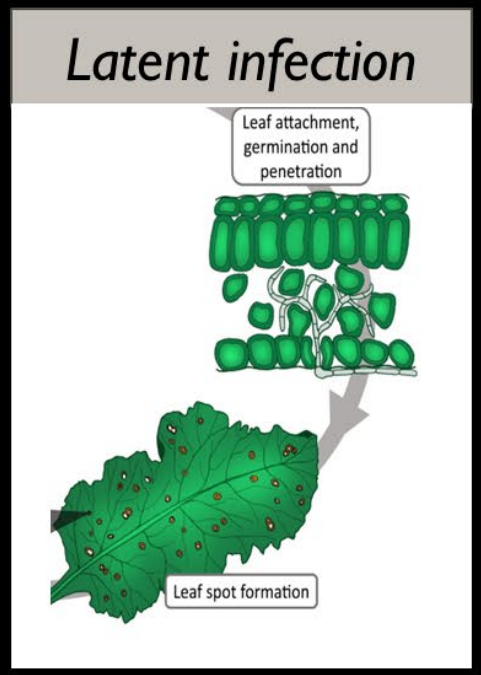
What is latent CLS disease?

When does latent CLS disease begin?



Results from Secor Lab, NDSU Plant Pathology

Latent infection



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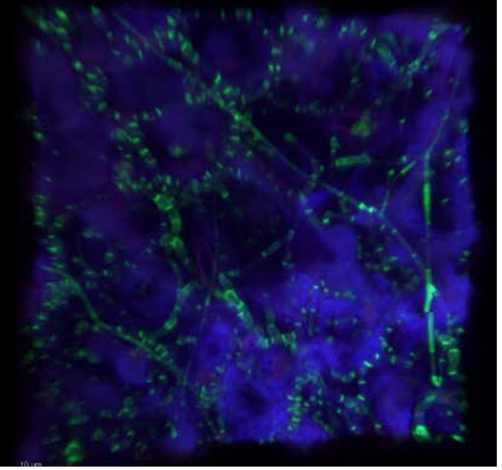
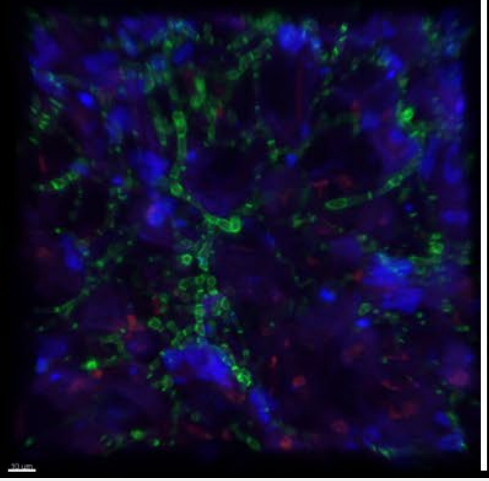


RED: DNA

BLUE: Cellulose

Green: Chitin

10 DPI



Intact plant nuclei

BLUE: Plant Cells

Green: Fungal Cells

Latent infection screening

Setup

With the help of ACSC, MinnDak, SMBSC:

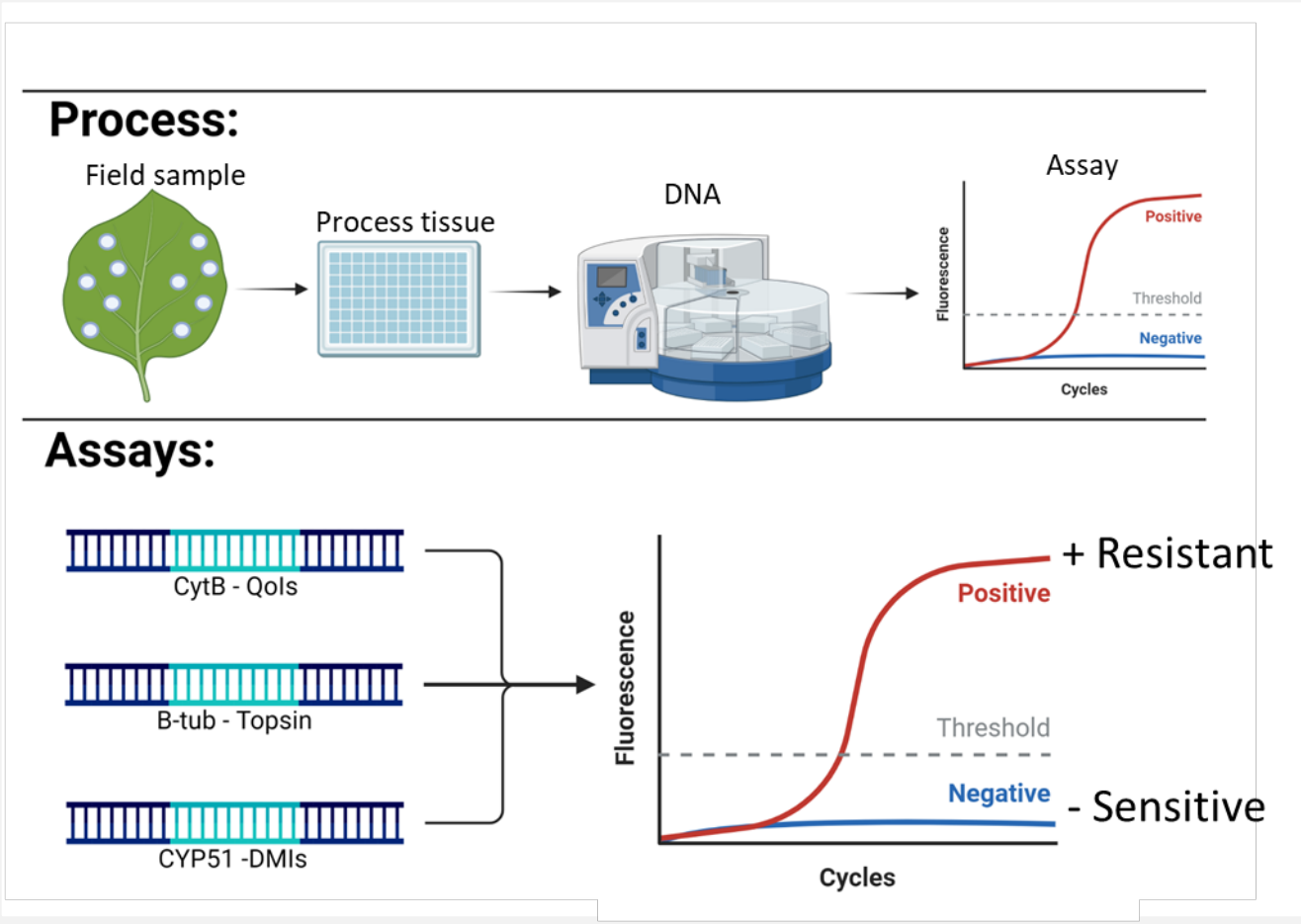
Sample **280** commercial fields from across the RRV weekly.

Multiplex qPCR assay:

QoI R/S (G143A)
DMI R/S (E170,L144F)
Benzimidazole R (E198A)

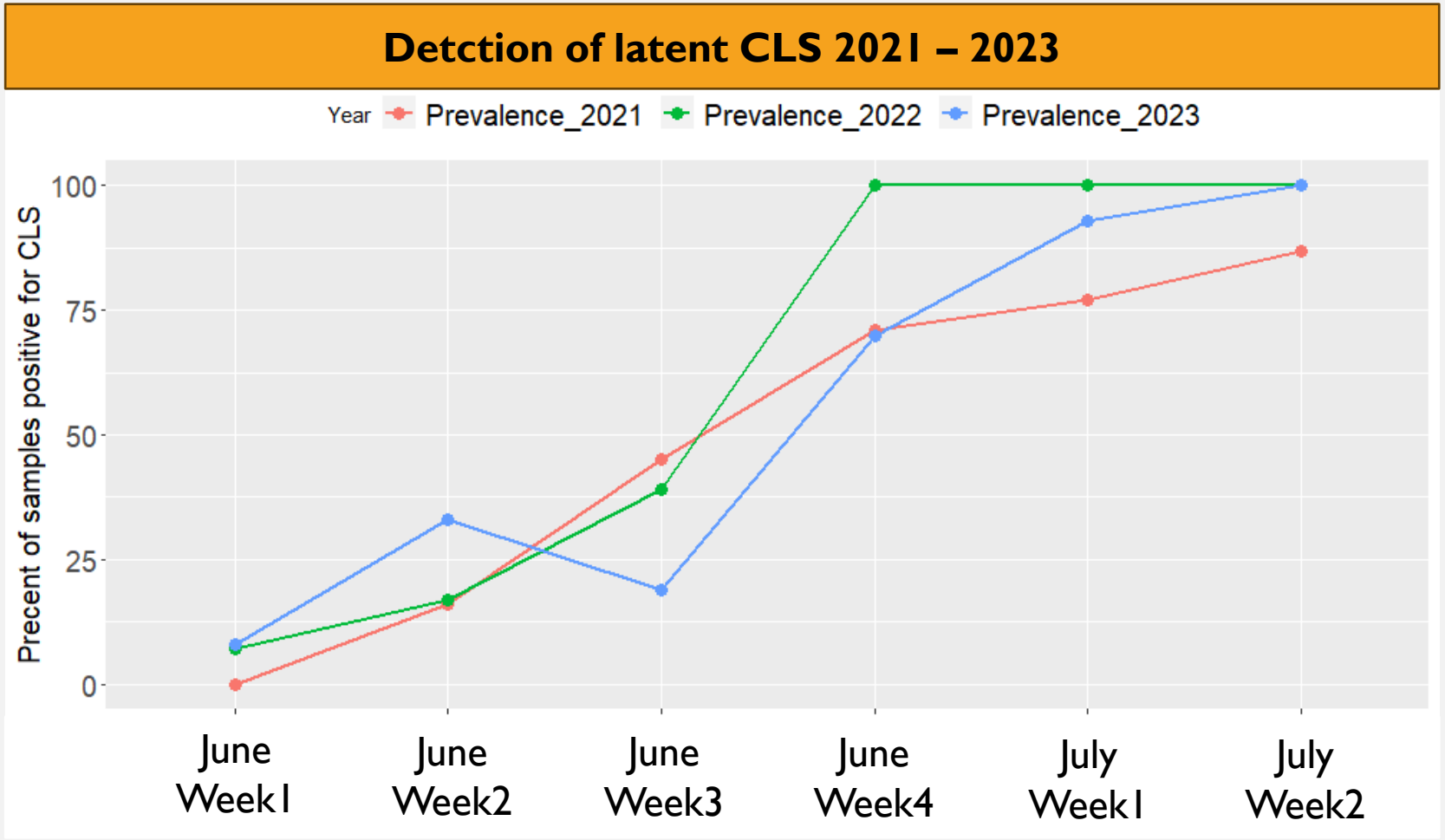
Report weekly results.

2 day turn around for 90 samples



Detecting *C. beticola* DNA in asymptomatic leaves.

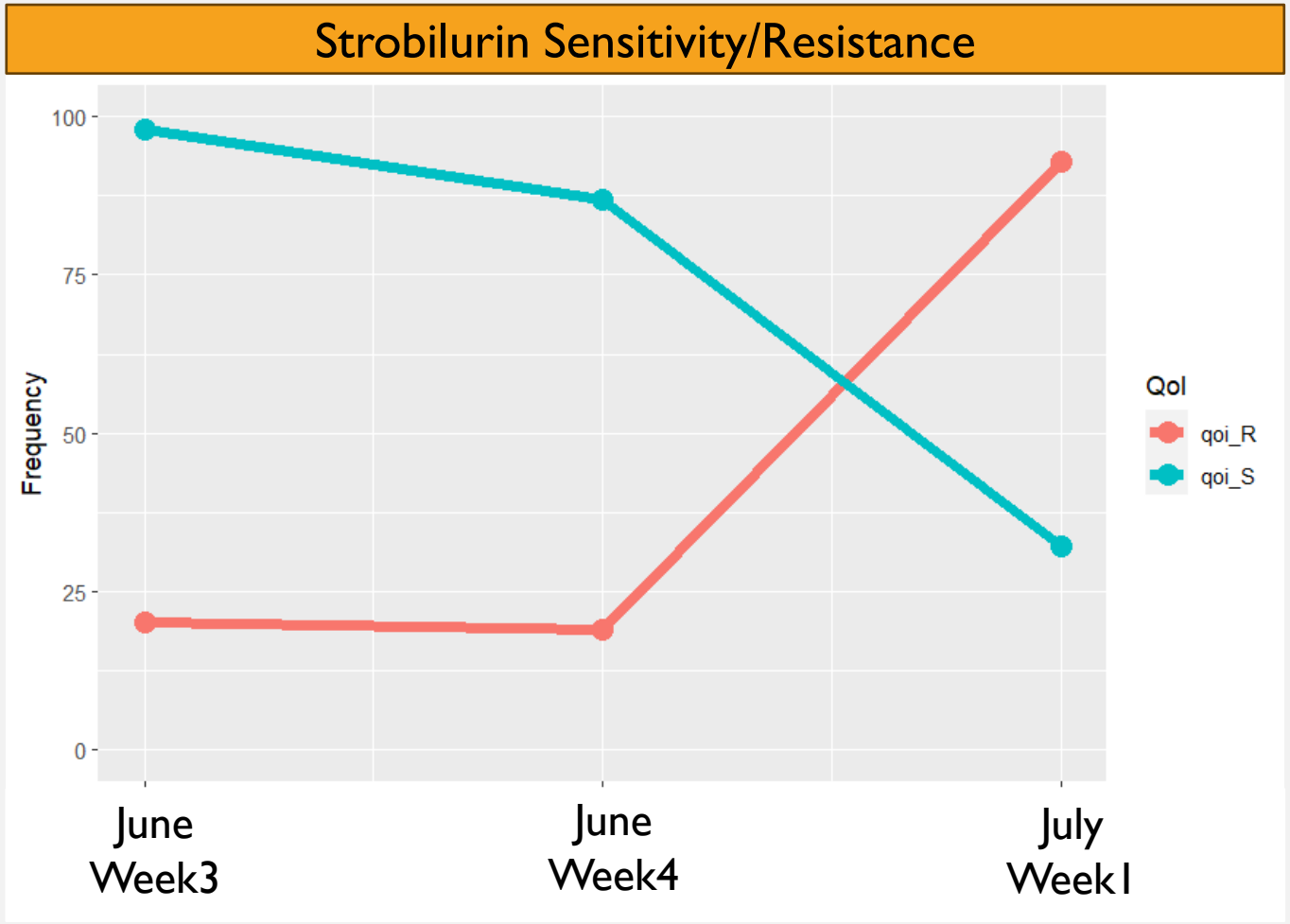
2021-2023 Latent CLS prevalence



Near **100%** of submitted samples are **positive** for latent CLS infection by the **first week of July** (~row closure).

Time to symptom development variable.

2023 Latent CLS Fungicide Resistance

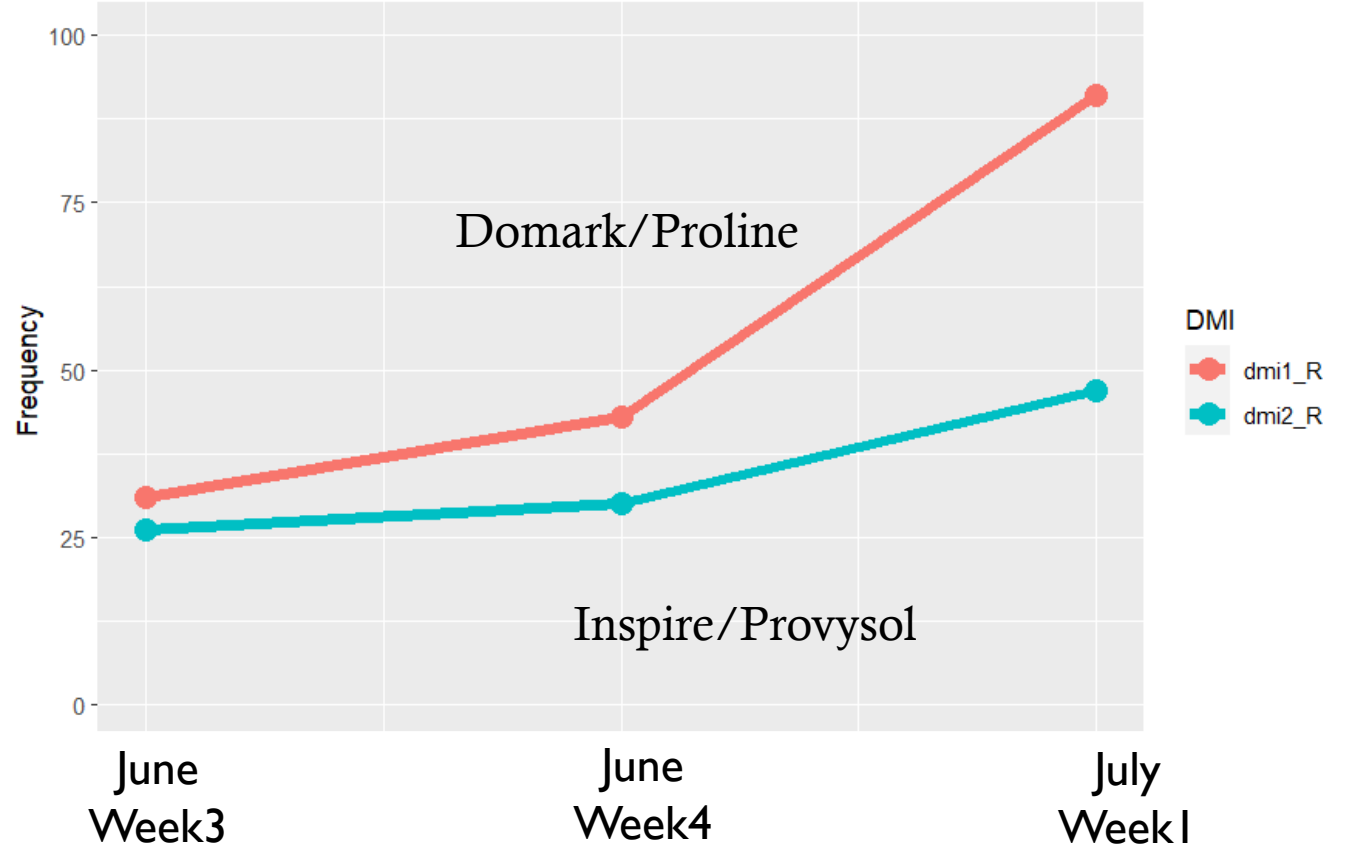


Early latent infection was mostly sensitive.

Late surge in Strobilurin resistance.

2023 Latent CLS Fungicide Resistance

Triazole Resistance

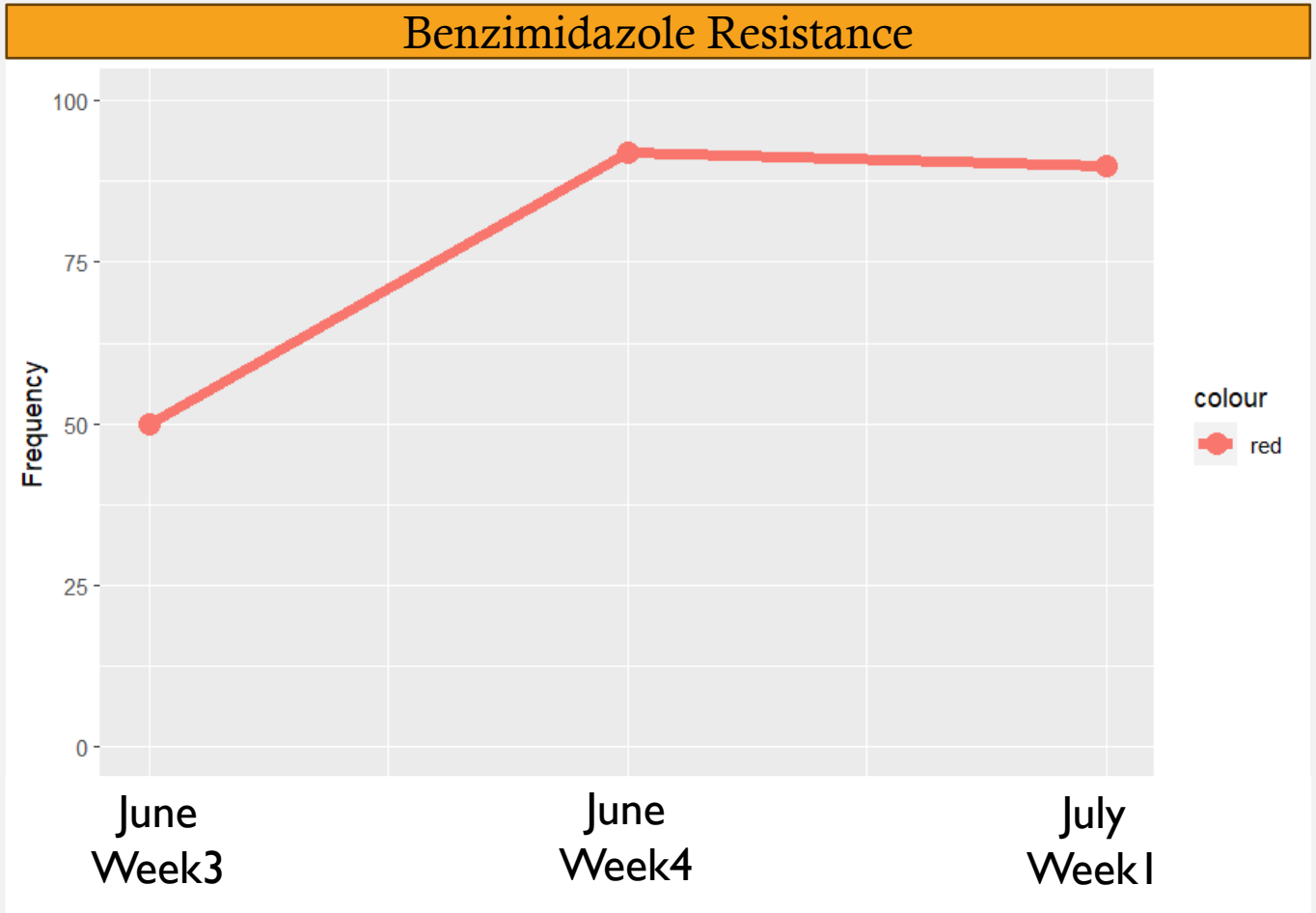


DMI resistance begins low and steadily rises.

Domark and Proline (red) resistance higher than Inspire and Provysol resistance (blue)

*Note we are not screening for sensitivity.

2023 Latent CLS Fungicide Resistance



Benzimidazole (Topsin) resistance begins low and steadily rises.

*Note we are not checking for sensitivity

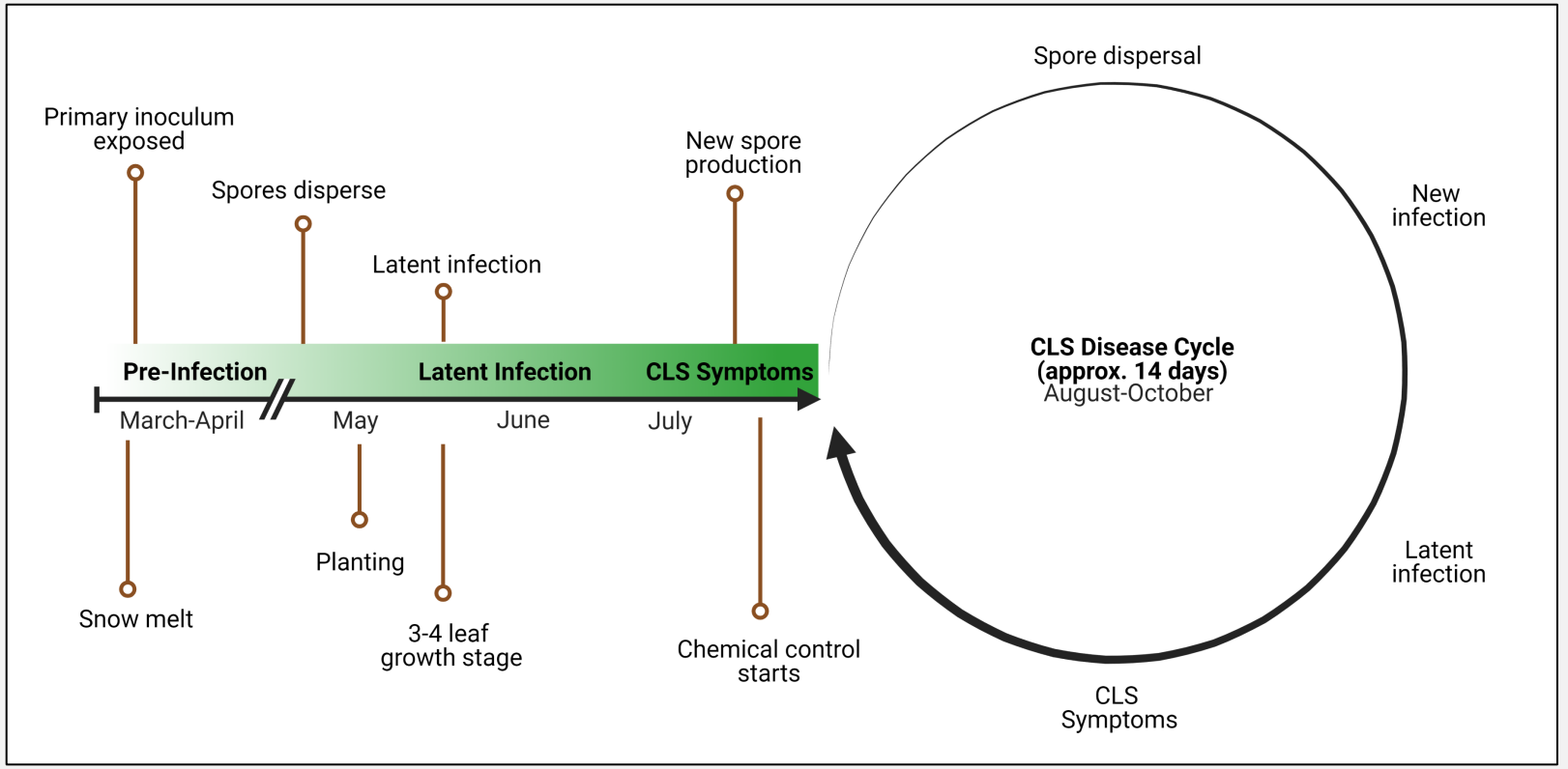
CLS disease cycle

When are spores dispersed in spring?
Detected beginning of May.

What conditions are favorable for spore germination?
Temperatures > 50 degrees F
Available free water (Rain/Dew)

What is latent CLS disease?
C. beticola infection before CLS symptoms

When does latent CLS disease begin?
Late June – Early July

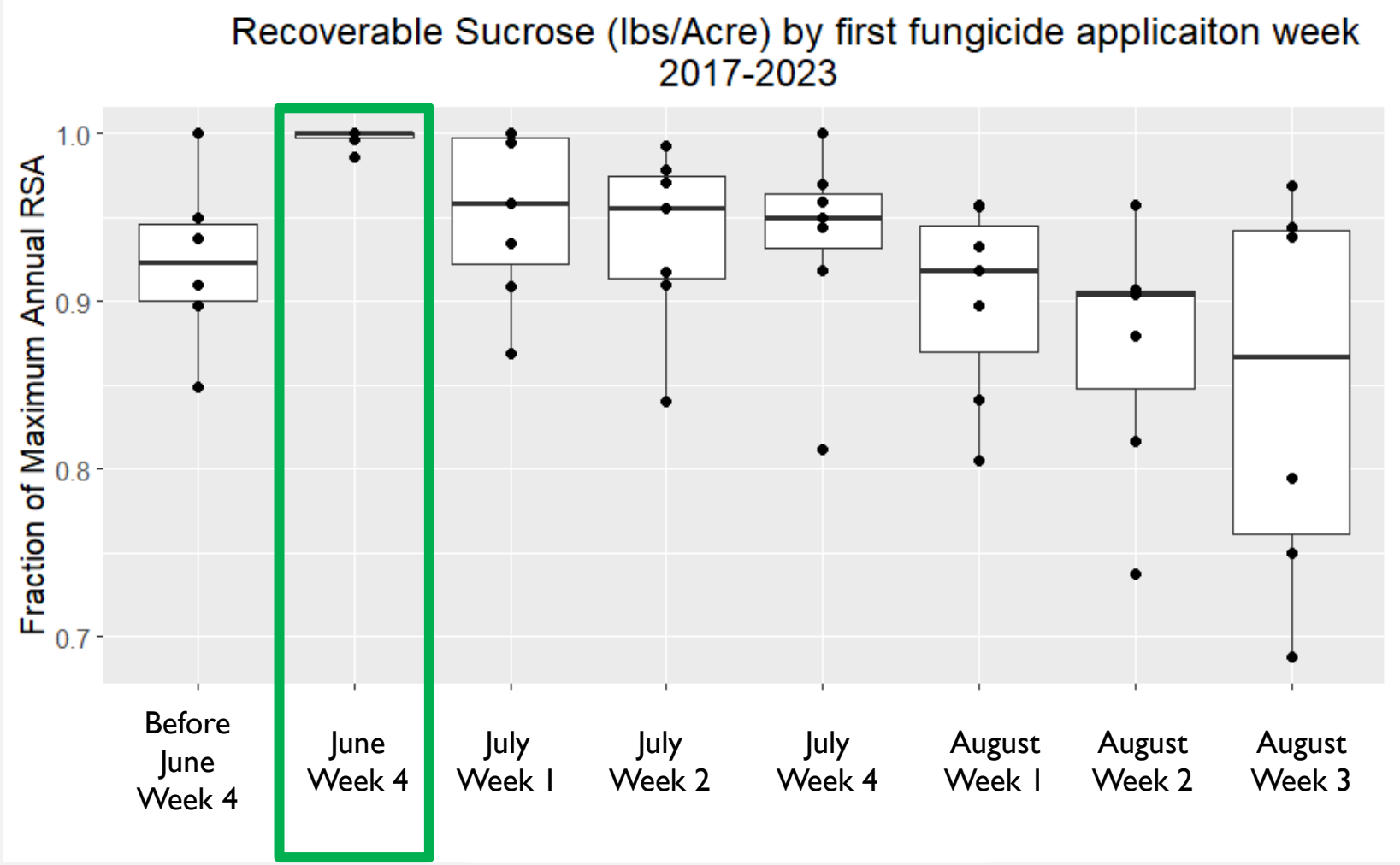


Why do we care if there are no symptoms?

Primary results

- Highest average annual RSA obtained when CLS fungicide control began in the last week of June.
- ~4% RSA reduction from June Week 4 to July Week 1

- Fields were organized by the start week of CLS control.
- The highest RSA week for each year was set to 1.0.
- All weeks were compared to the best week within the year to make comparisons.

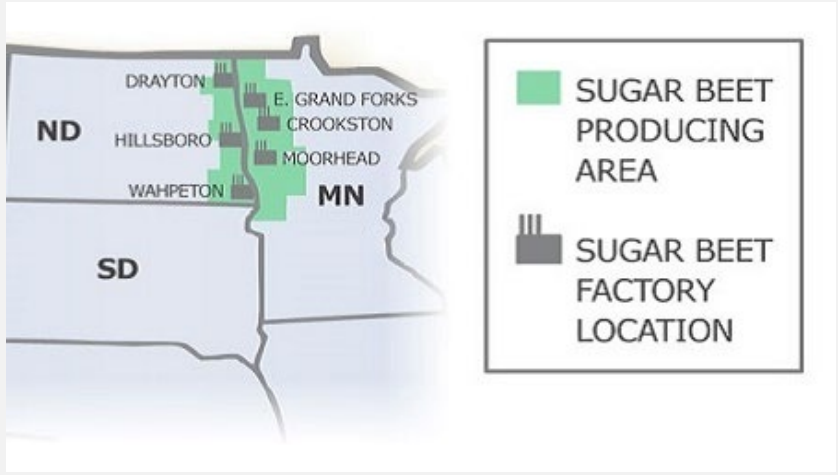


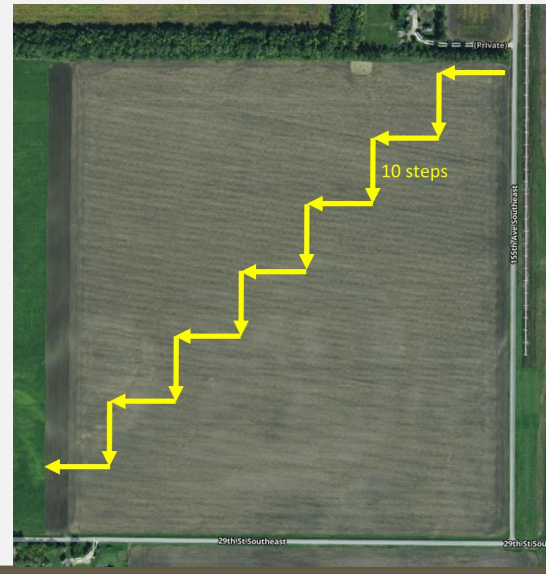
Shorter Timescales: In Season Dynamics

What affect to fungicide treatments have on the pathogen population throughout the season?

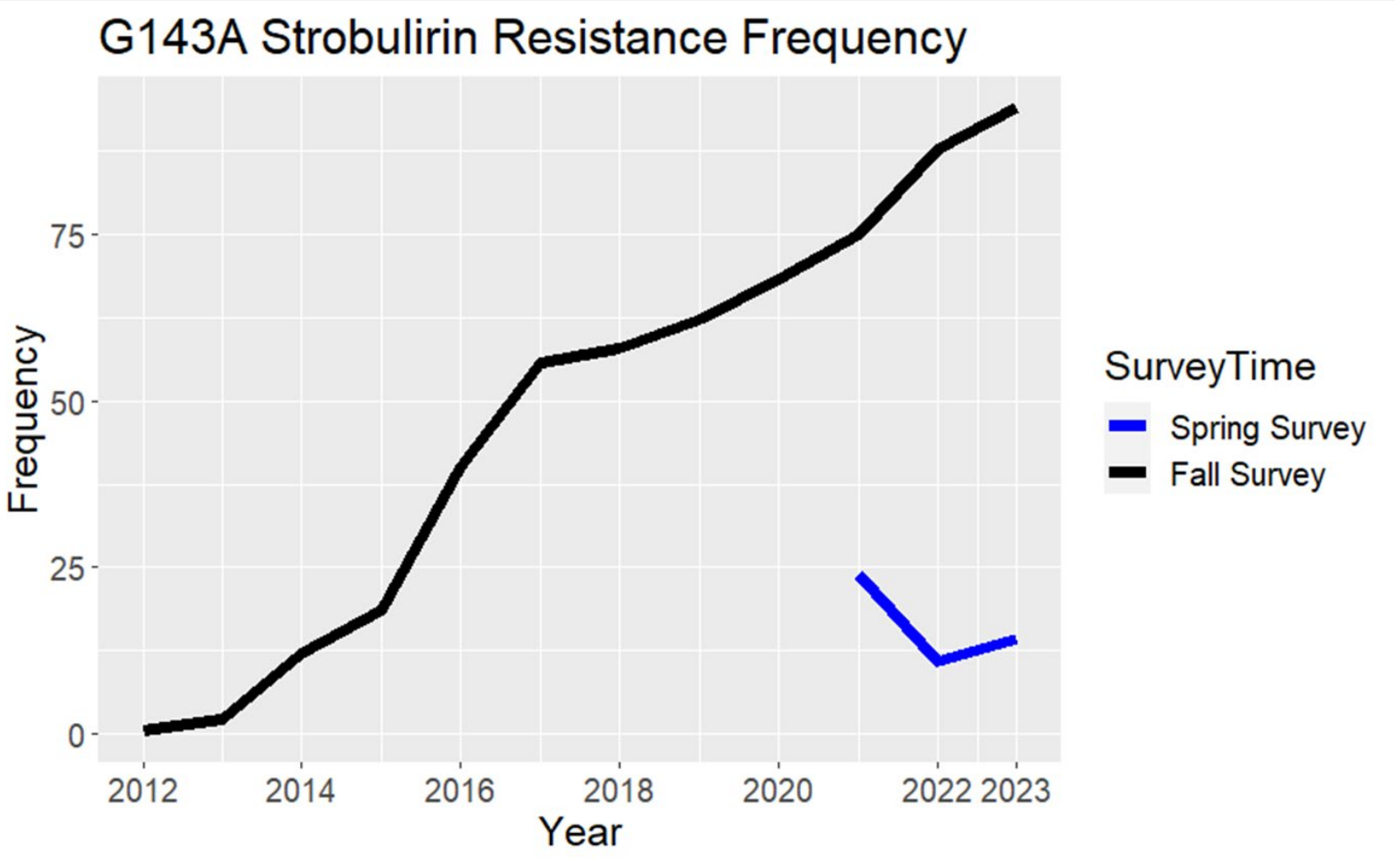
Are there differences in the amount of fungicide resistance detected in a field when sampled at different times of the year?

Samples taken before any fungicide application and after each subsequent application.





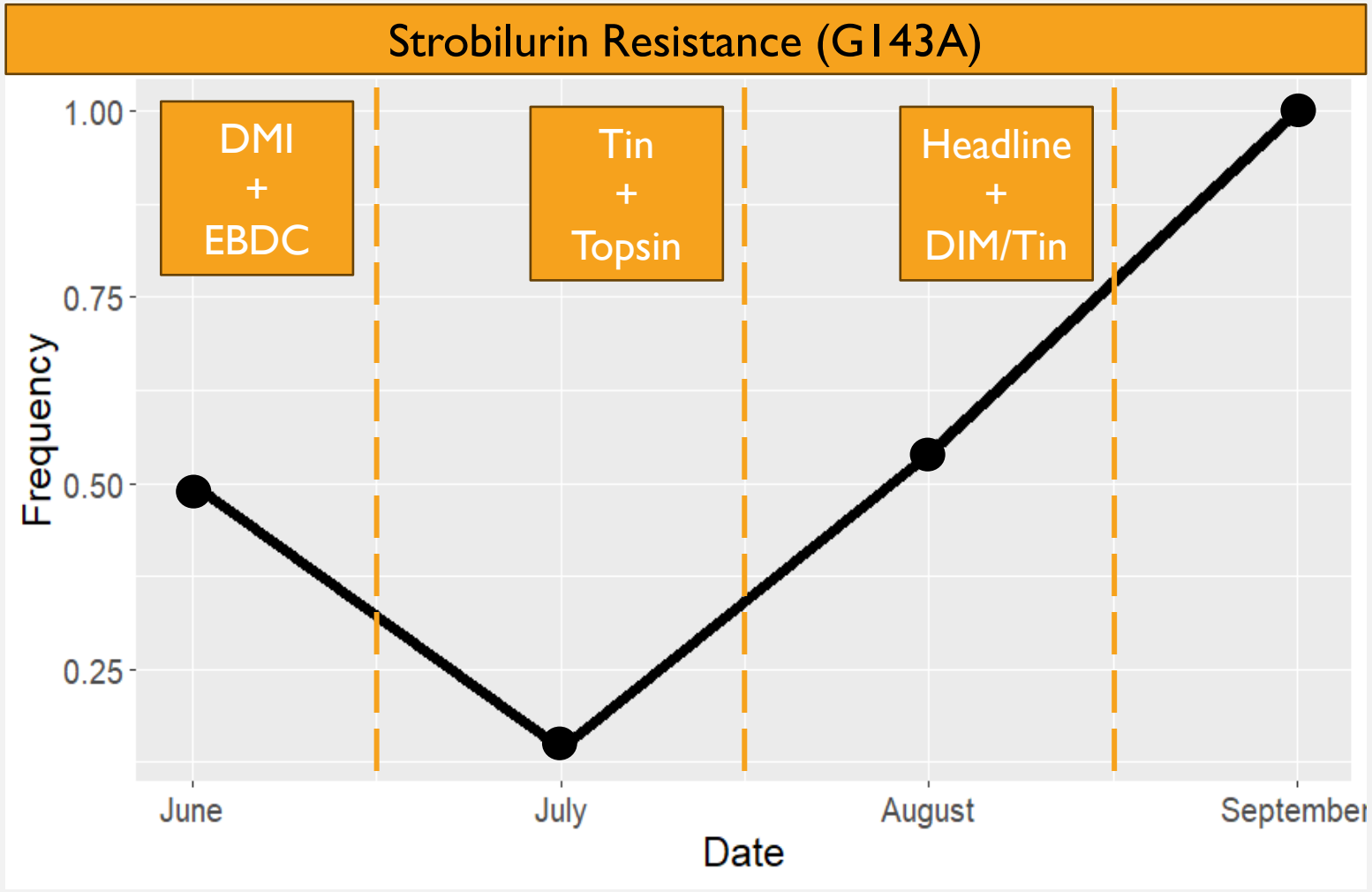
Annual Strobilurin Resistance Fluctuations



Primary results

- Strobilurin resistance frequency is lower in the spring
- Spring survey based on Spore trap data and Latent infection data
- Fall surveys detect high frequencies of Strobilurin resistance

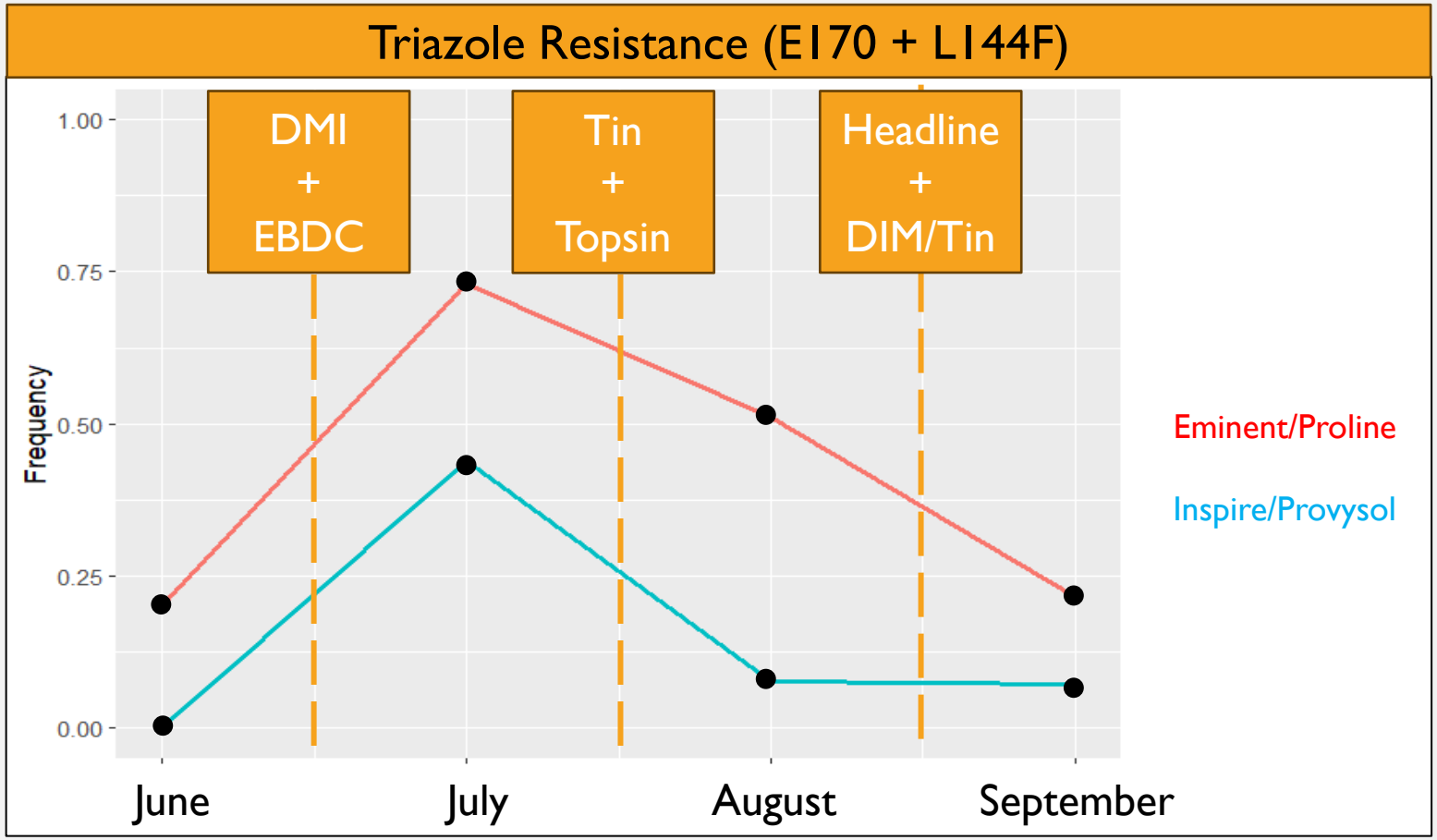
Annual Strobilurin Resistance Fluctuation



Primary results

- Strobilurin resistance changes throughout the year in response to management practices
- Specifically, we observe a decline in Strobilurin resistance following Triazole applications.
- Data based on 8 fields in 2023 and 3 fields in 2022

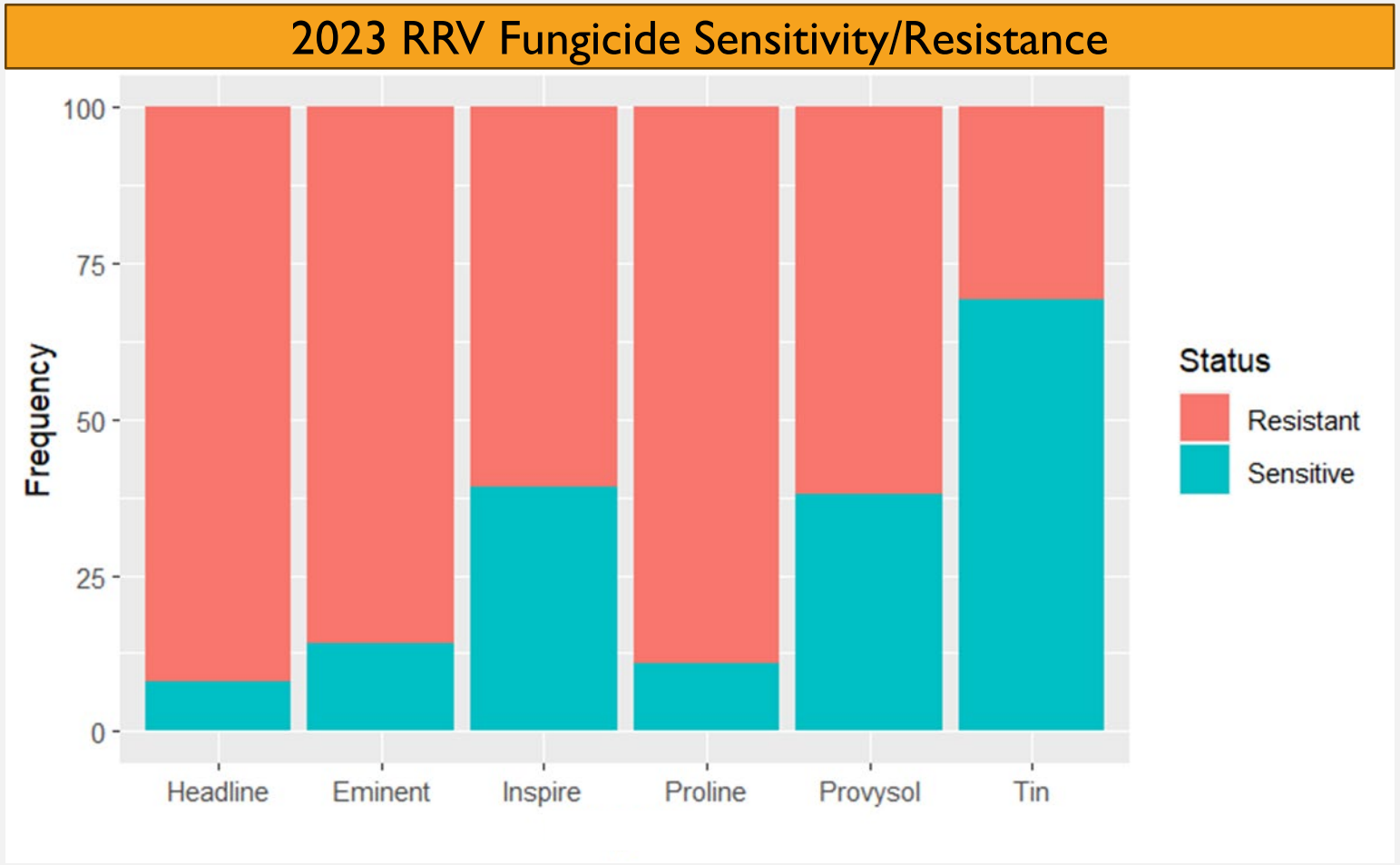
Annual Triazole Resistance Fluctuation



Primary results

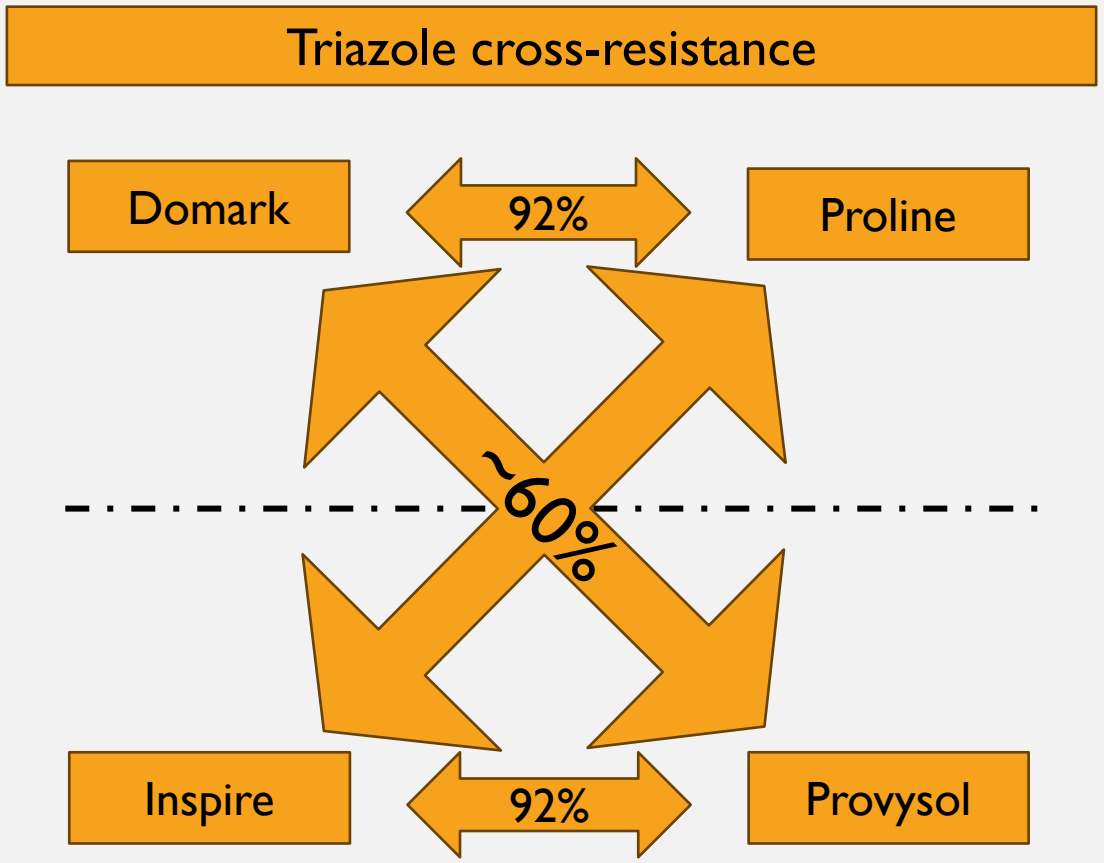
- Triazole resistance changes throughout the year in response to management practices
- Data based on 8 fields in 2023 and 3 fields in 2022
- Note that increased detection of Triazole resistance in September resulted if Triazole was also applied (2 fields).

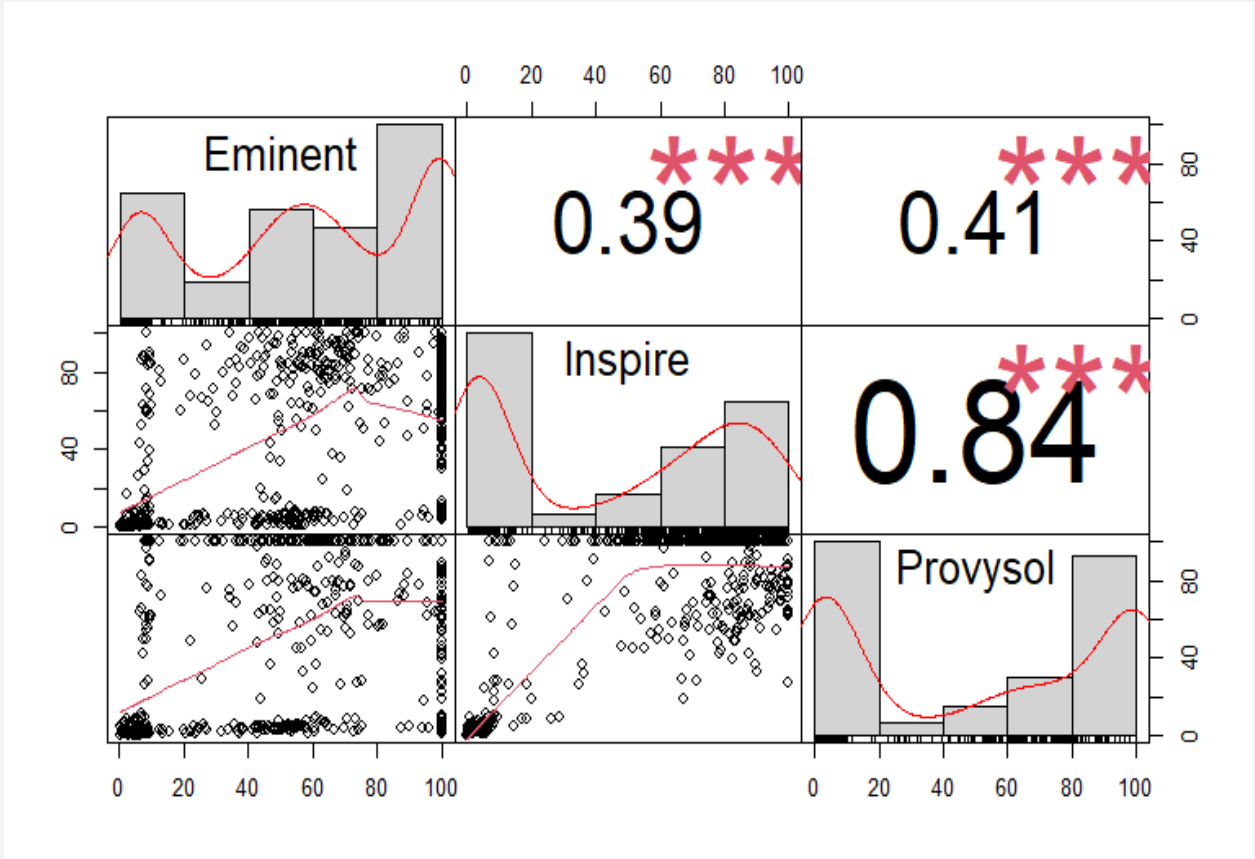
Fungicide resistance 2023



- Headline resistance frequency is high at the End of the Year (EOY) – 92%
- Triazole resistance frequency is high at the EOY
  - Eminent: 86%
  - Proline: 89%
  - Inspire: 61%
  - Provysol: 62%
- Tin resistance is lower than previous years:
  - 2023: 31%
  - 2022: ~98%
  - 2021: ~95%

2023 Fungicide Cross Resistance – RRV



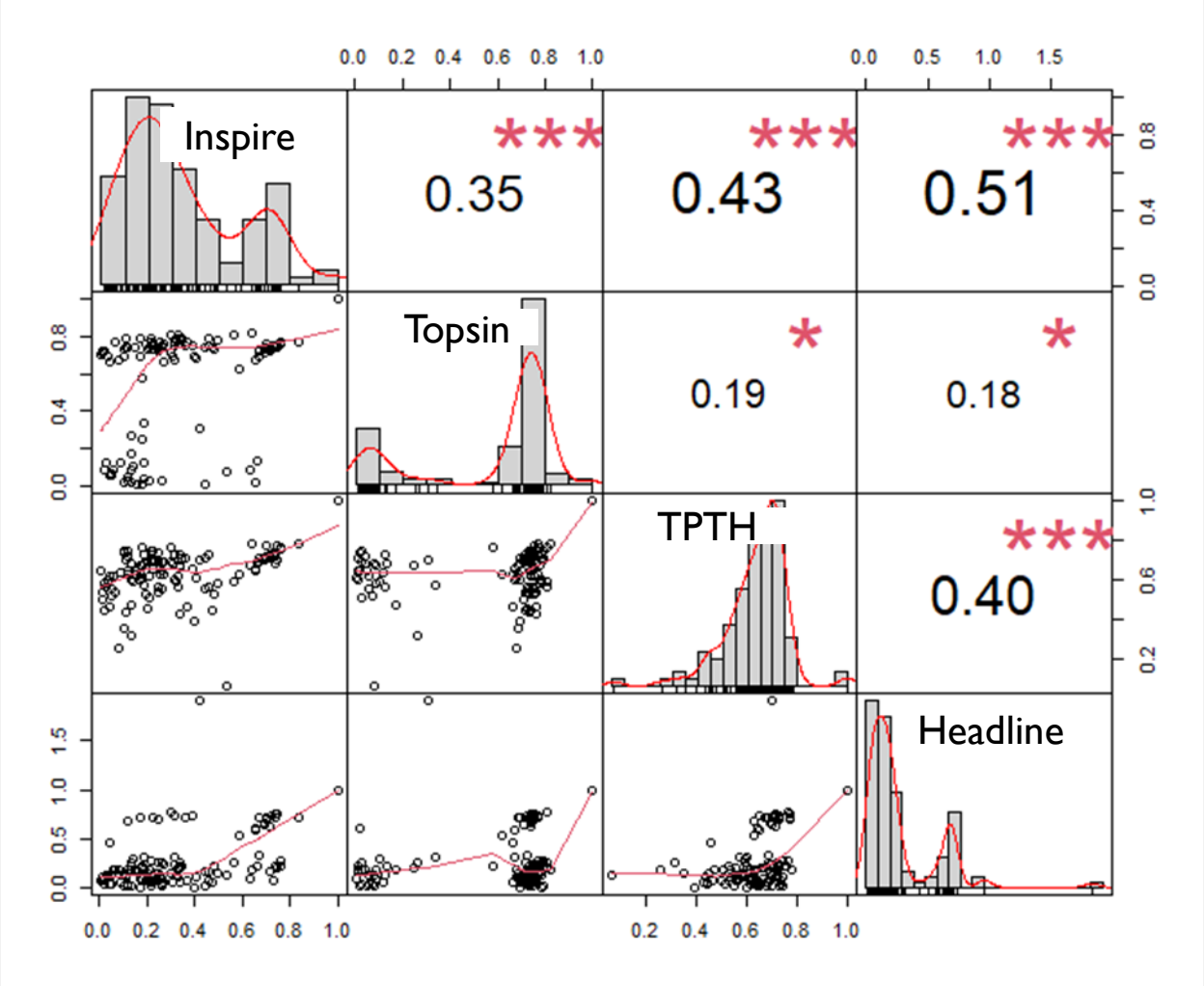


Cross resistance between Triazole compounds is not always present.

2023 Fungicide Cross Resistance – RRV

Cross-resistance

- Low cross resistance between different chemistries
  - Between Triazoles with cross resistance we observe correlation coefficients >0.84 out of a potential high score of 1.0
  - Between different chemistries the correlations are much lower (0.18-0.51)
- The lack of cross resistance indicates that effective CLS control should incorporate:
  - **Tank Mixes**
  - **Rotating chemistries within the season**



Take aways

- CLS latent infection prevalence reached ~100% at the beginning of July in 2021-2023 and corresponds relatively well with row closure.

- Molecular assays can be used to detect latent infection as well as detect fungicide resistance mutations.

- Fungicide resistance is not fixed, management practices influence development and maintenance of resistance in the *Cercospora beticola* population.

- Cross resistance is low between fungicide chemistries but can be high within a chemistry.
  - DMI cross resistance falls into two categories with Domark/Proline and Inspire/Provysol
  - Rotate chemistries
  - Tank mix

Acknowledgements

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Sugarbeet Research & Education Board

BEET SUGAR DEVELOPMENT FOUNDATION

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