Molecular epidemiology of Cercospora leaf spot

Fungicide resistance and host adaptation in the Red River Valley

2025 Grower Seminar

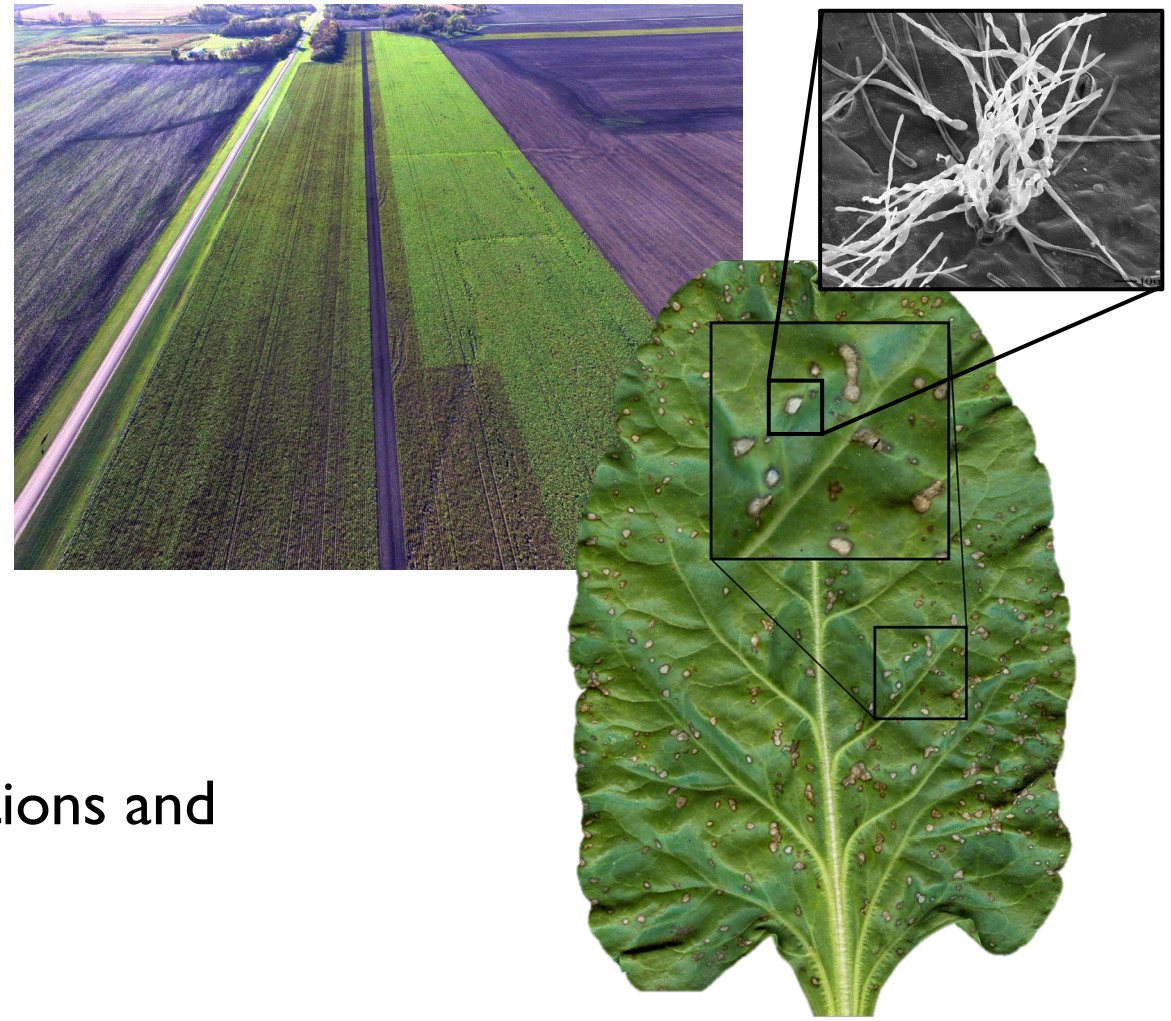
February 6th 2025 – Grand Forks, ND

February 11th 2025 – Fargo, ND

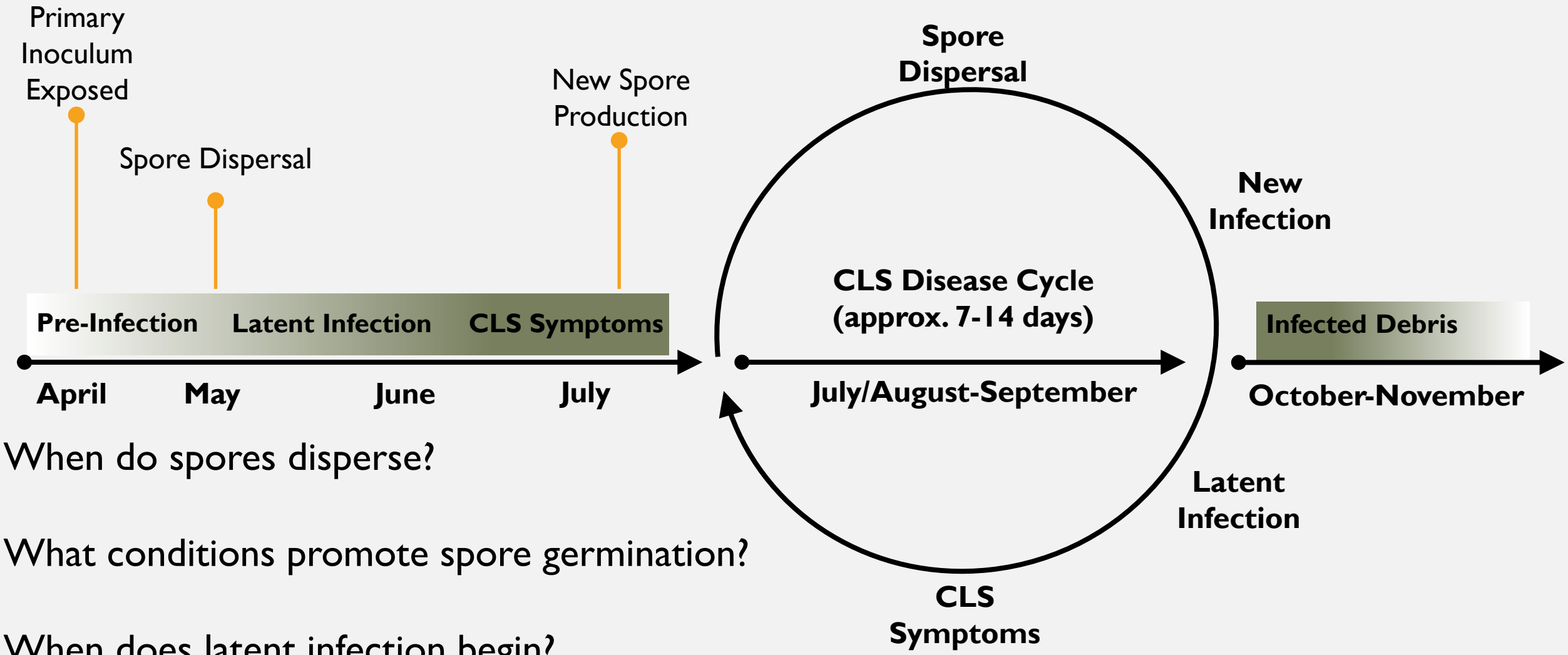
February 13th 2025 – Grafton, ND

Cercospora beticola

- Cercospora leaf spot (CLS) on sugarbeet
- Hemibiotrophic fungus
  - Asymptomatic biotrophic phase
  - Symptomatic necrotrophic phase
- Polycyclic and genetically diverse
  - Cryptic sexual cycle
- Primarily controlled through fungicide applications and resistant sugarbeet varieties.



CLS disease cycle

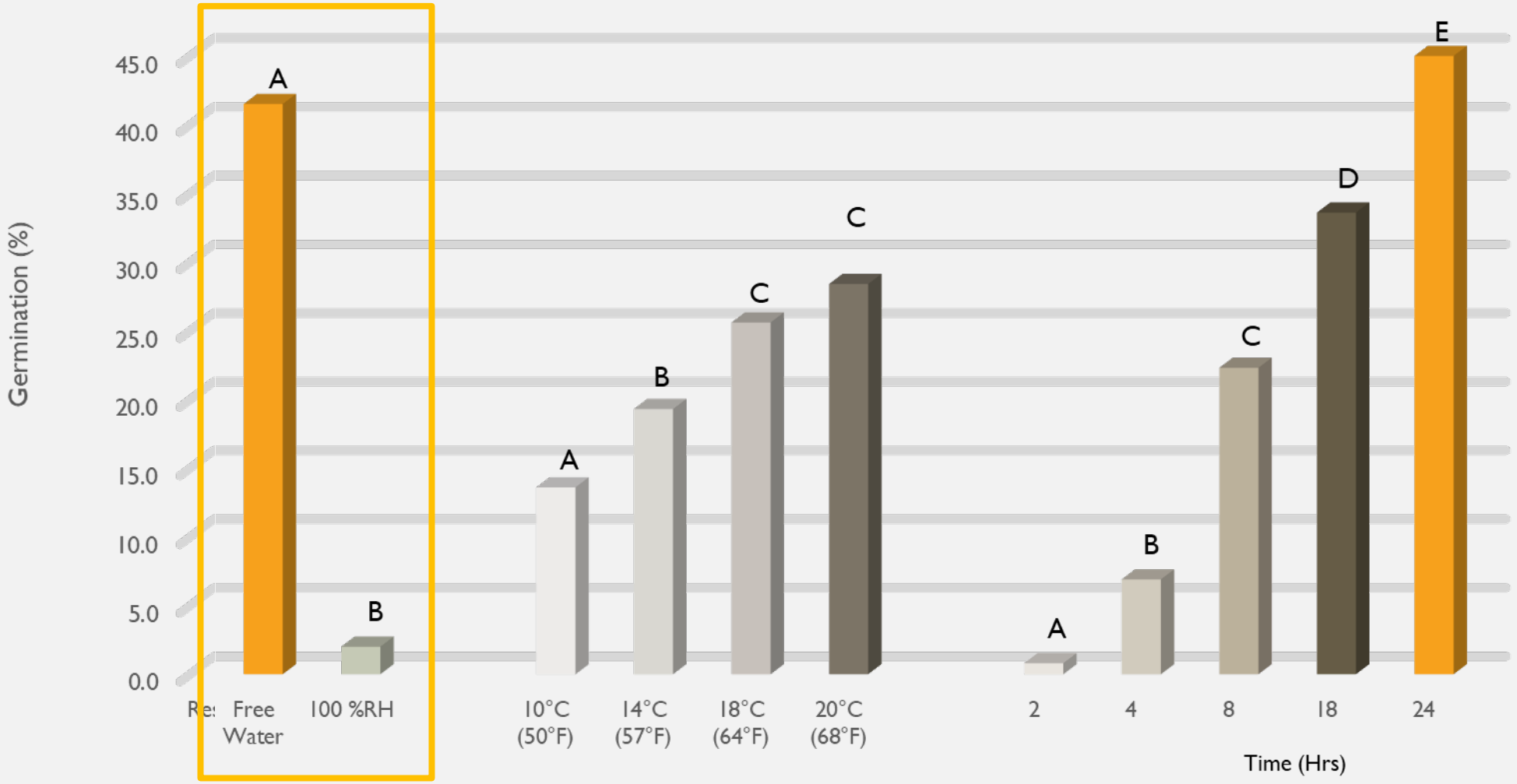


When do spores disperse?

What conditions promote spore germination?

When does latent infection begin?

C. beticola spore germination



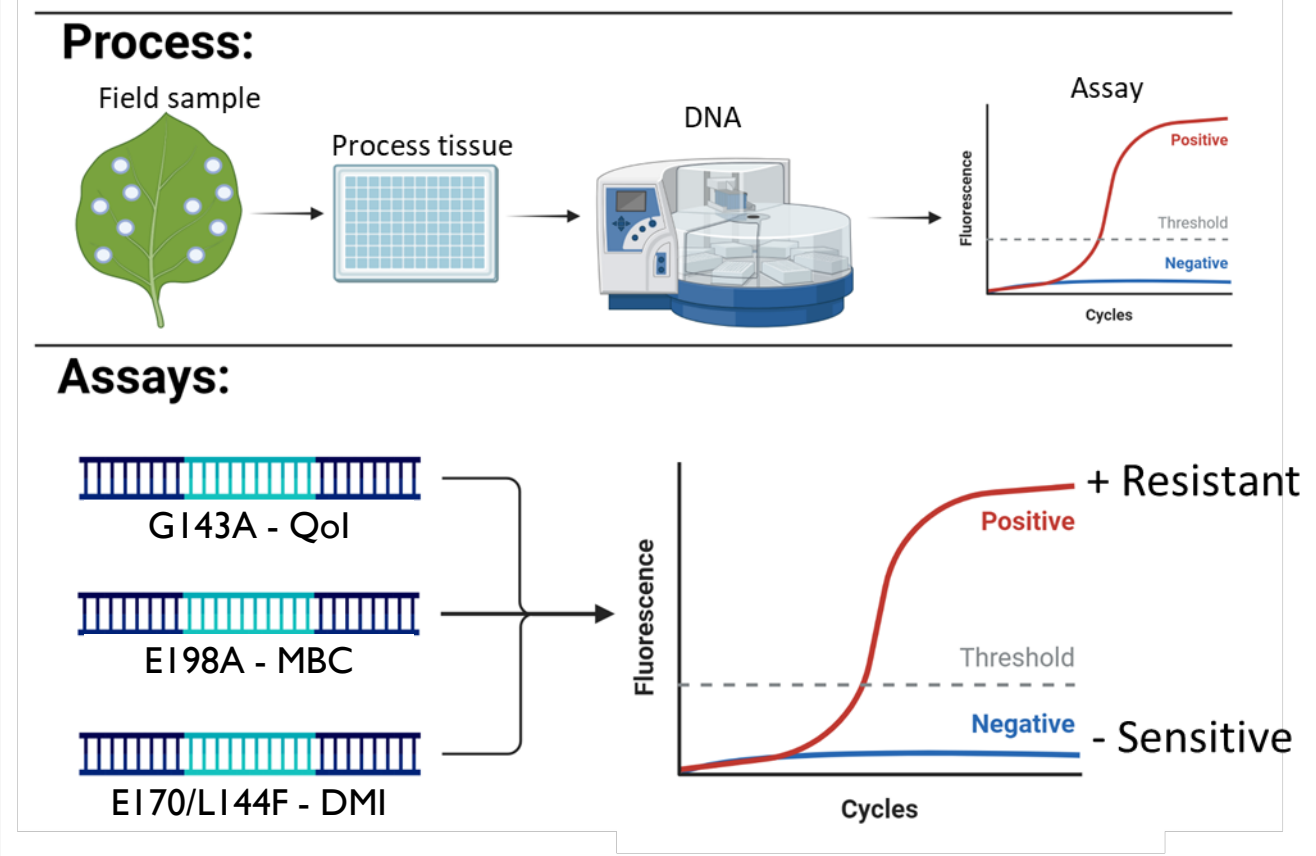
Data Provided by Dr. Gary Secor

Latent infection screening

Survey of commercial sugarbeet fields

- Conducted in 2021 – 2024
- 280 commercial sugarbeet fields
  - Weekly sampling starting at the 4-6 leaf stage

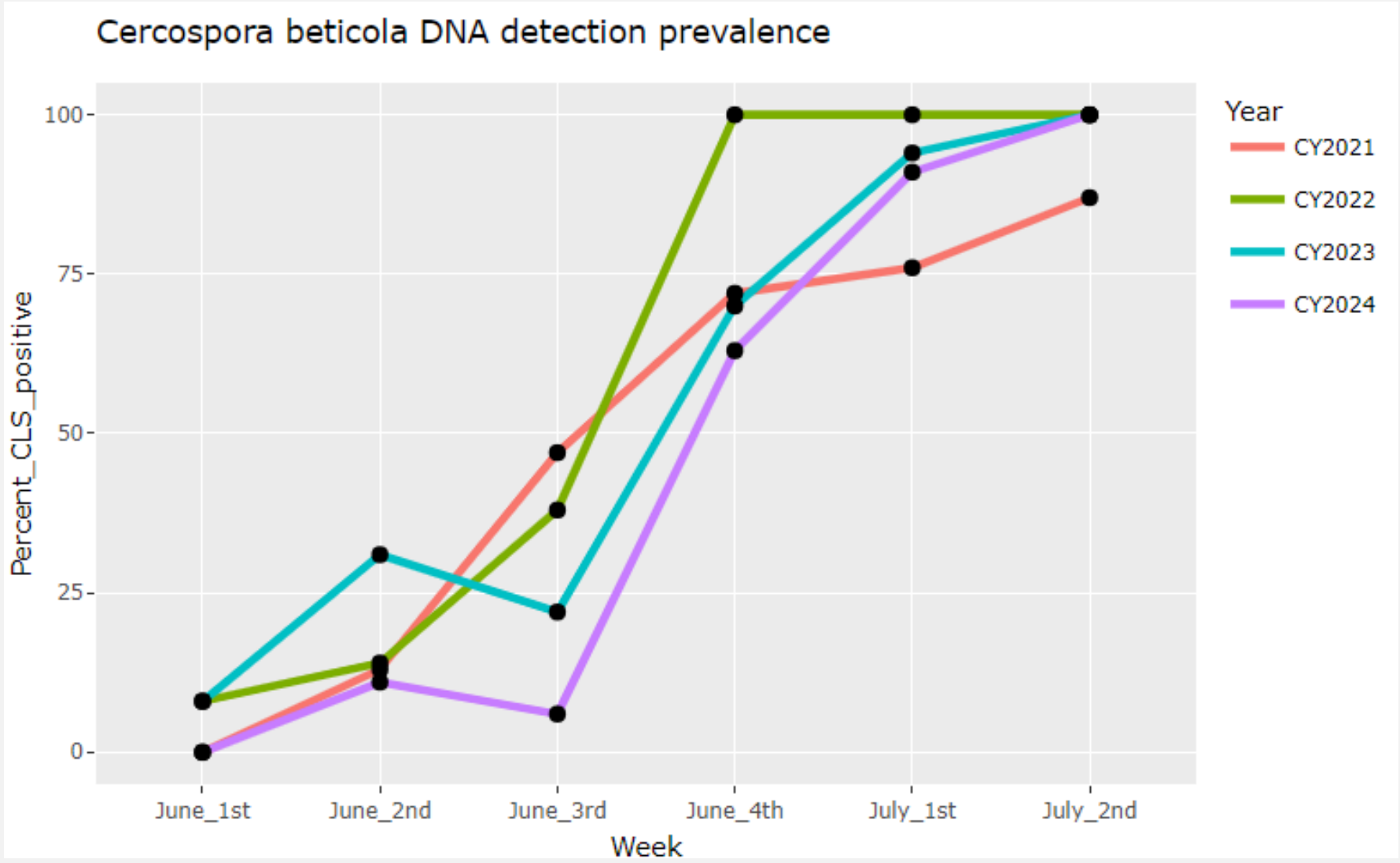
- Multiplex assay
  - Bolton Lab USDA
  - G143A – QoI marker
    - wildtype and mutant
  - E198A – MBC marker
  - E170/L144F – DMI markers



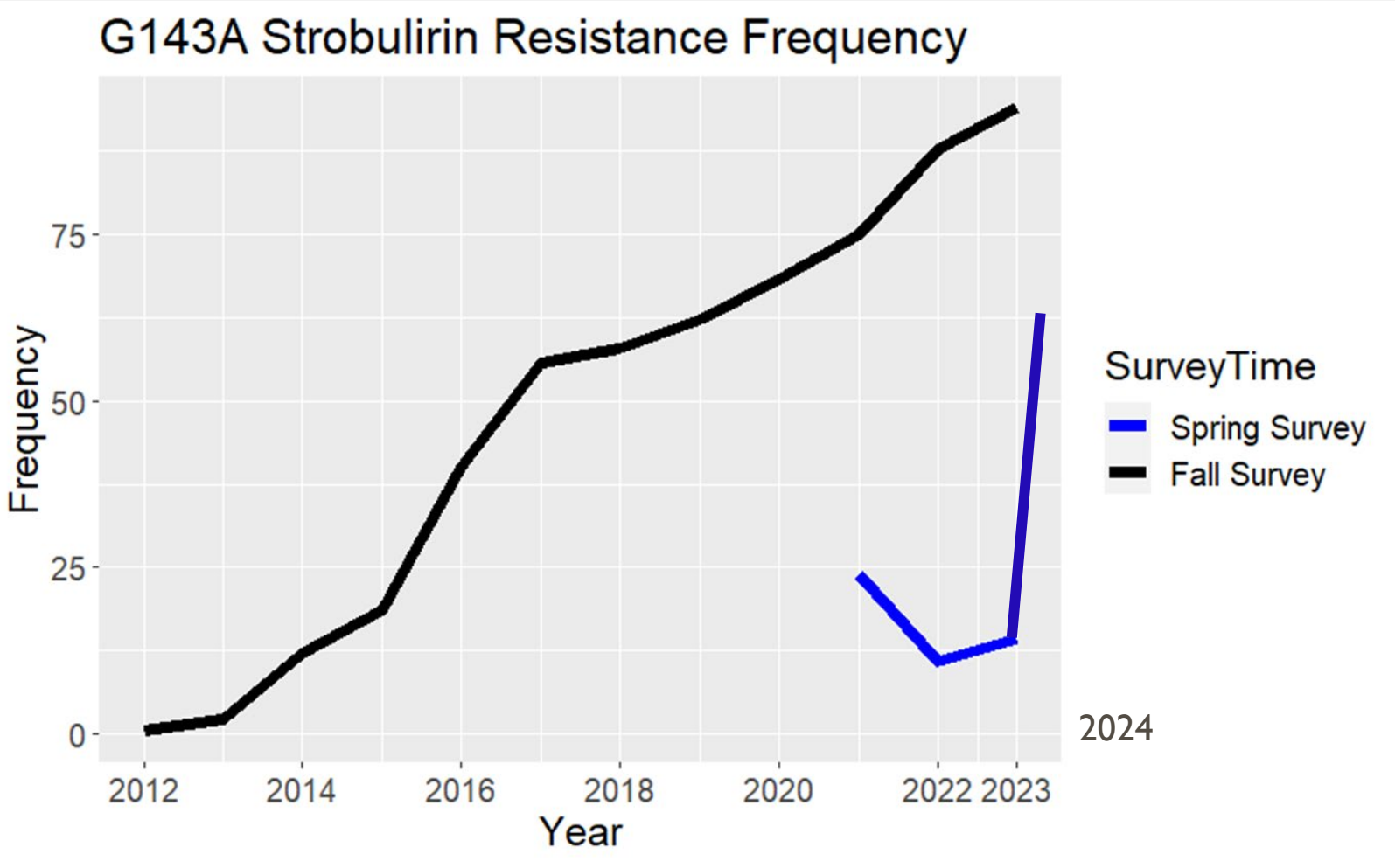
2021-2024 Latent CLS prevalence

Primary results

- Latent infection onset consistently occurs at or just prior to row closure in the RRV



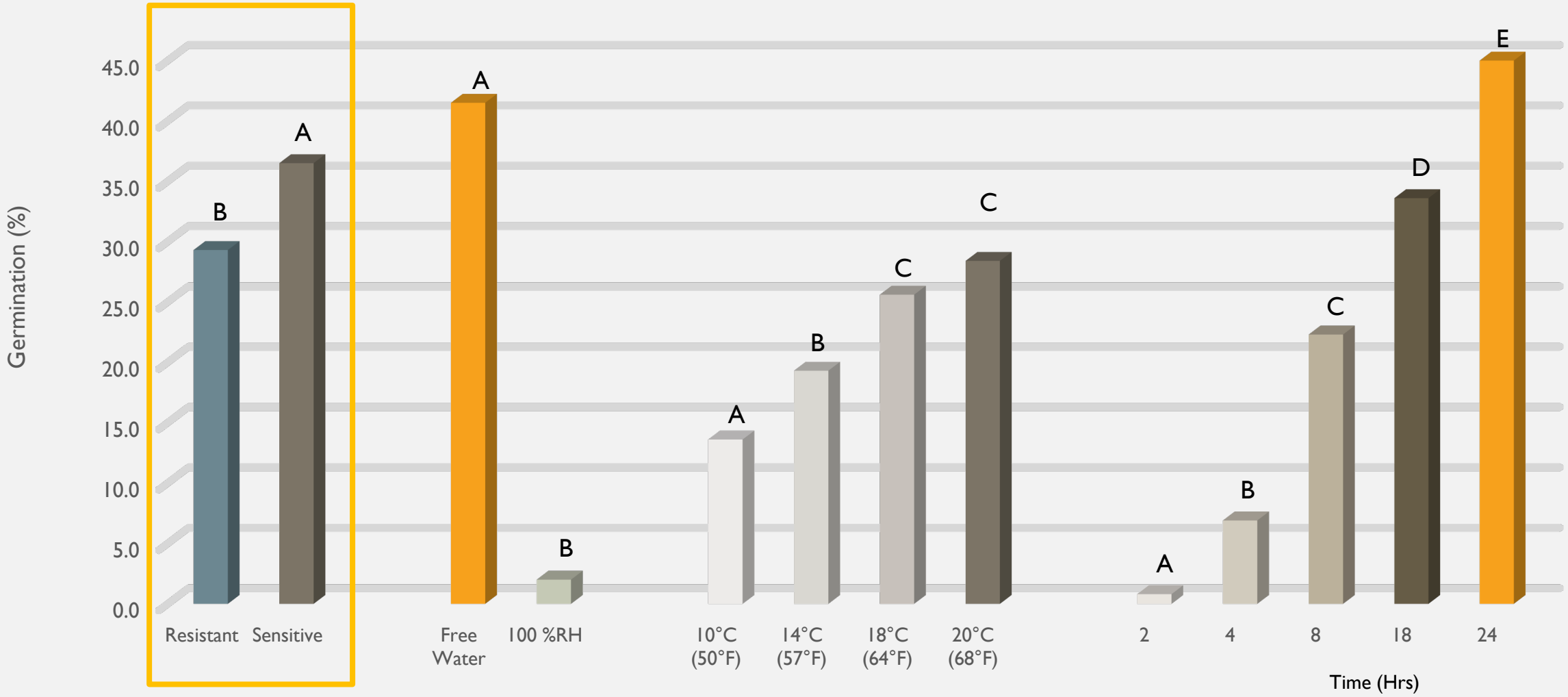
Annual Strobilurin Resistance Fluctuations



Primary results

- Stobilurin resistance is lower in the spring
- Spring survey based on Spore trap data and Latent infection data
- What caused the increase in 2024 Strobilurin resistance in the spring

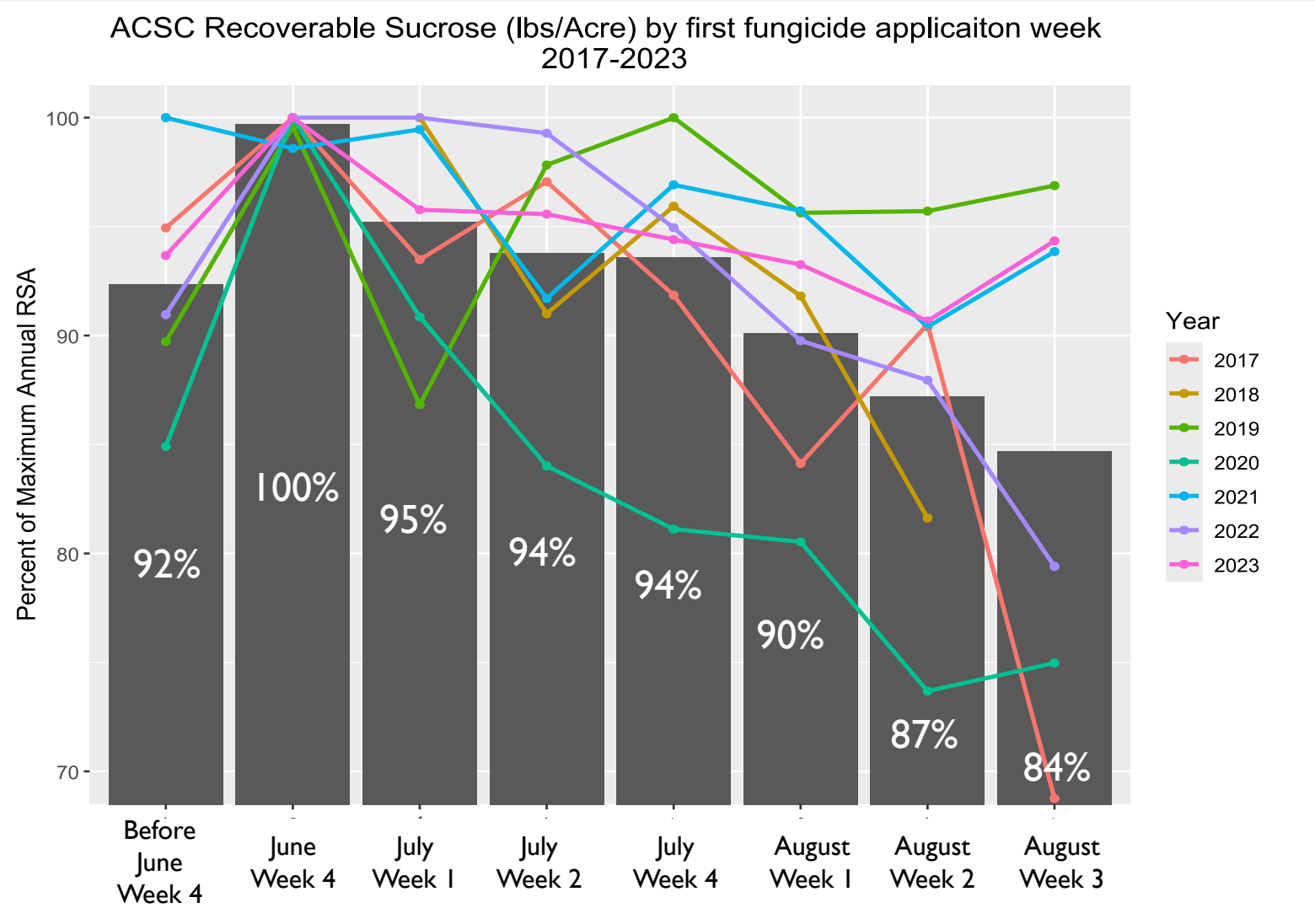
C. beticola spore germination



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.
- ~5% RSA reduction from June Week 4 to July Week 1
- 2019: Low CLS pressure year
- 2020: High CLS pressure year
- 2022: Low CLS pressure year



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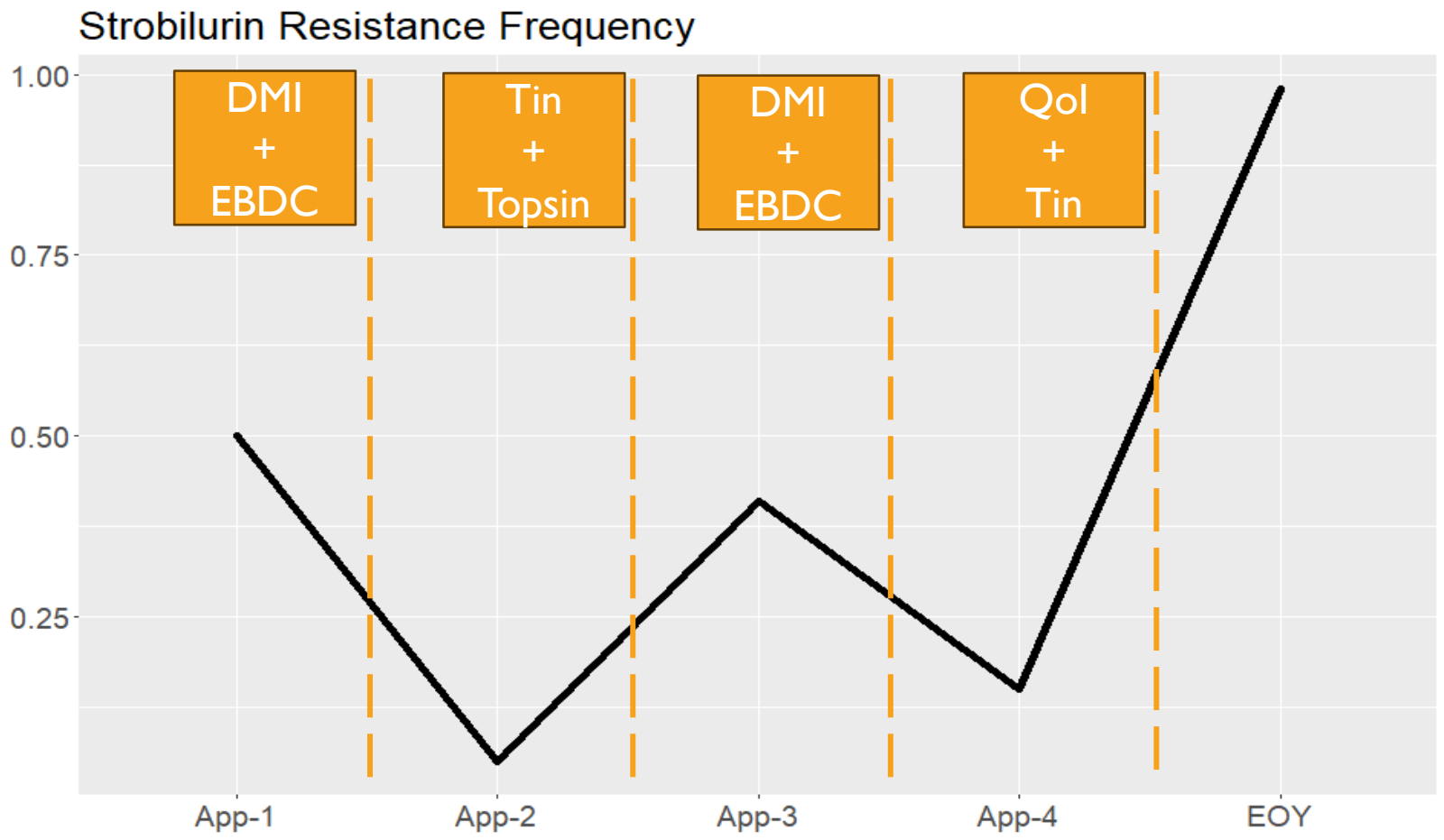
- 2019: Low CLS pressure year
- 2020: High CLS pressure year
- 2022: Low CLS pressure year

- Predicted missed potential between $21.1 to $104.1 million dollars.

Year	Acres	$Earned	$Potential	$Difference
2019	196,554	$242.6 M	$263.7 M	$21.1 M
2020	371,361	$569.2 M	$673.3 M	$104.1M
2021	388,718	$731.9 M	$766.1 M	$34.2 M
2022	399,566	$838.9 M	$918.2 M	$79.3 M
2023	400,145	$1,217.3 M	$1,286.2 M	$69.0 M

Annual Strobilurin Resistance Fluctuation



Primary results

- Strobilurin resistance changes throughout the year in response to management practices
- DMI applications show potential to reduce strobilurin resistance in the field.

Fungicide cross resistance

Fungicide	Commercial product	Headline	Topsin	Domark	Proline	Inspire	TPTH
Strobilurin	Headline	1.00					
Benzimidazole	Topsin	0.18	1.00				
Triazole	Domark	0.69	0.33	1.00			
Traizole	Proline	0.53	0.41	0.92	1.00		
Triazole	Inspire	0.51	0.37	0.59	0.60	1.00	
Tin	TPTH	0.40	0.21	0.48	0.54	0.43	1.00

- Cross resistance scored from 0.00 to 1.00 with higher values indicating higher degrees of cross resistance.

Primary results

- Cross resistance is relatively low between different chemistries
- Tank Mixing multiple chemistries as an effective strategy
- Currently recommended Tank Mixes (ACSC):

Fungicide cross resistance

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Strobilurin	Headline	1.00					
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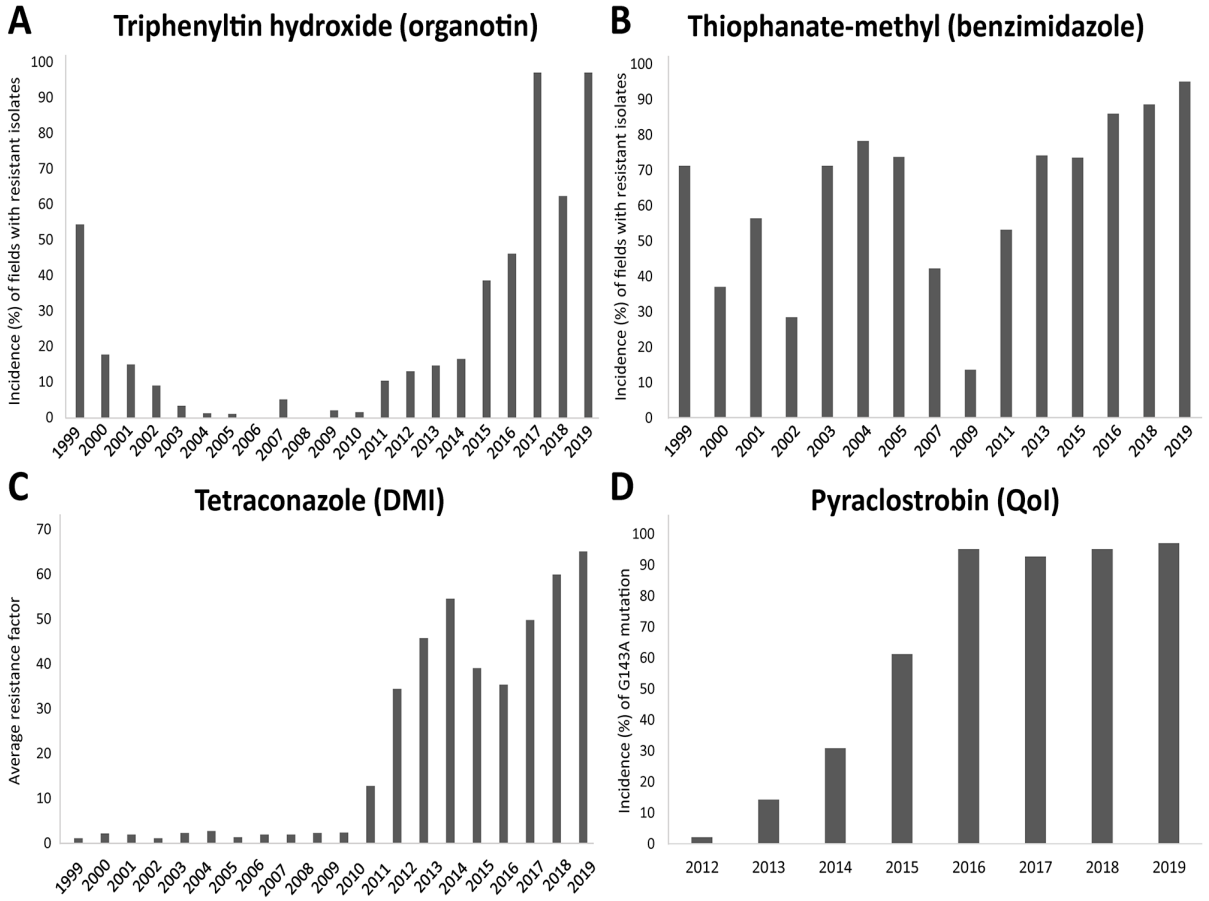
- Cross resistance scored from 0.00 to 1.00 with higher values indicating higher degrees of cross resistance.

Primary results

- Cross resistance is relatively low between different chemistries
- Tank Mixing multiple chemistries as an effective strategy
- No EBDC resistance has been detected.

C. beticola population dynamics



Fungicide	2016	2017	2021	2023
Strobilurin	29%	31%	20%	25%
Triazole	81%	46%	72%	62%
Triazole	47%	45%	51%	53%
Benzimidazole	30%	14%	23%	15%
Organotin	31%	22%	35%	30%

There are differences between the frequency of resistance detected on a field or regional basis and on a per isolate/individual basis

C. beticola population dynamics

Fungicide	Commercial name	Mutation	2016	2017	2021	2023
Strobilurin	Headline	G143A	29%	31%	20%	25%
Triazole	Domark/Proline	E170	81%	46%	72%	62%
Triazole	Inspire/Provysol	L144F	47%	45%	51%	53%
Benzimidazole	Topsin	E198A	30%	14%	23%	15%
Organotin	SuperTin	GST	31%	22%	35%	30%

Frequency of fungicide resistance mutations in whole genome sequenced *C. beticola* isolates collected at the end of season survey.

Though the incidence of fungicide resistance is high across fields for any level of resistance, the individuals in those fields show lower resistance levels.

Example: Most fields sampled in 2021 had isolates that were Tin resistant.

BUT not all of the isolates in any one field were resistant.

C. beticola population dynamics

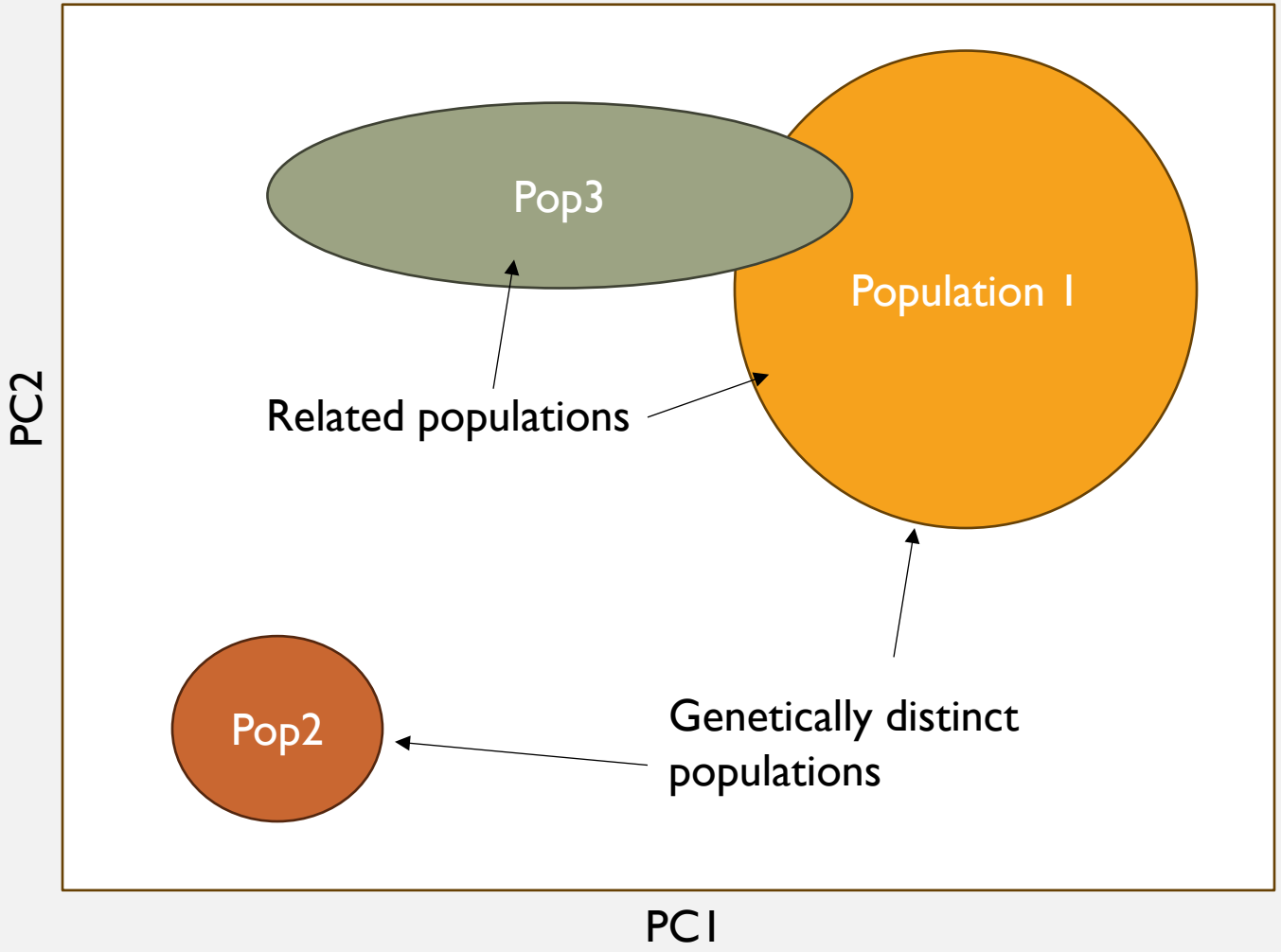
WGS and PCA Primer

Whole genome sequencing of RRV populations to identify mutations.

Principle component analysis can be used to identify patterns of genetic variation among individuals/populations.

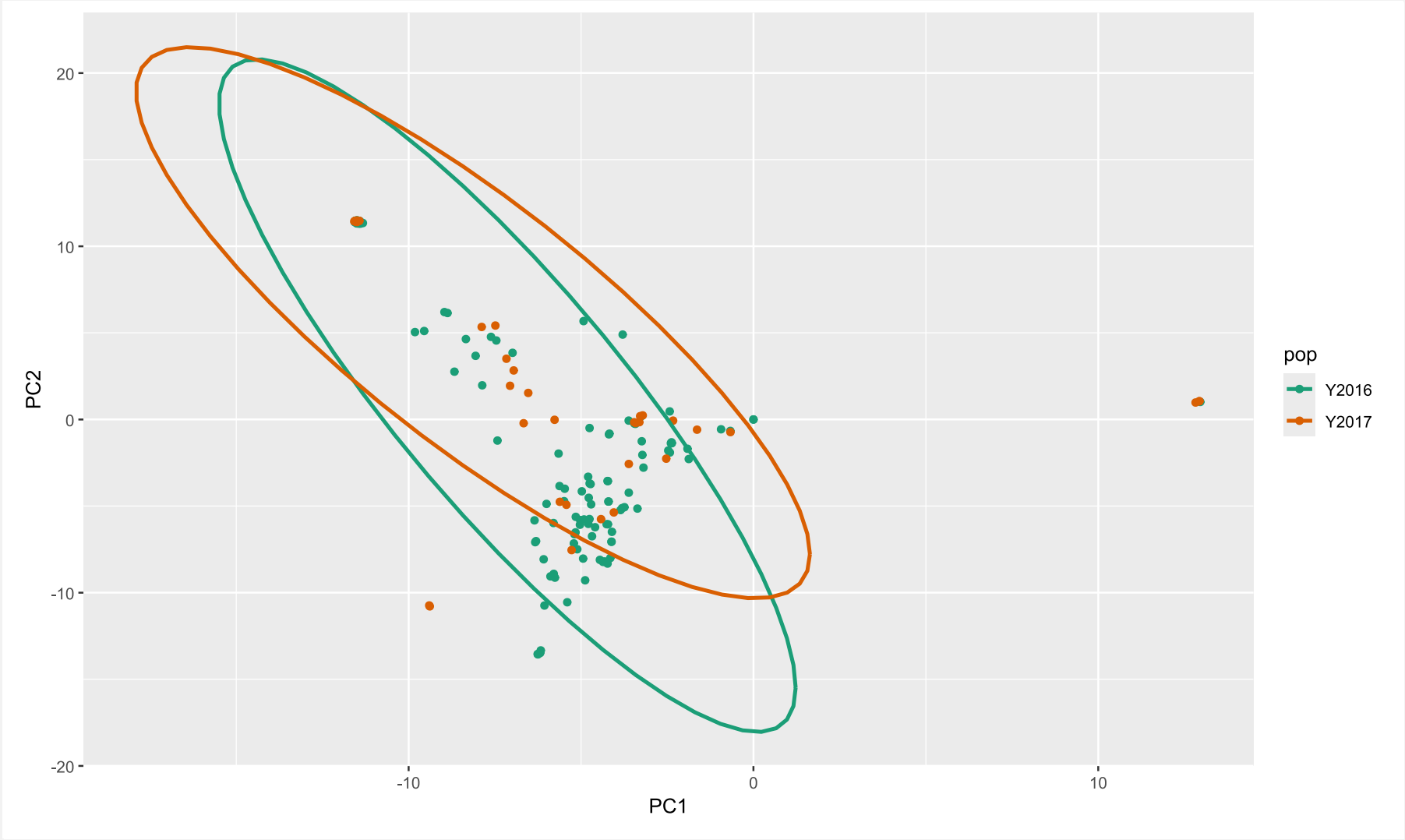
Factors commonly influencing population structure:

- Geography
- Sampling timeline
- Environmental conditions
- Specific selective pressures
  - i.e. Management practices



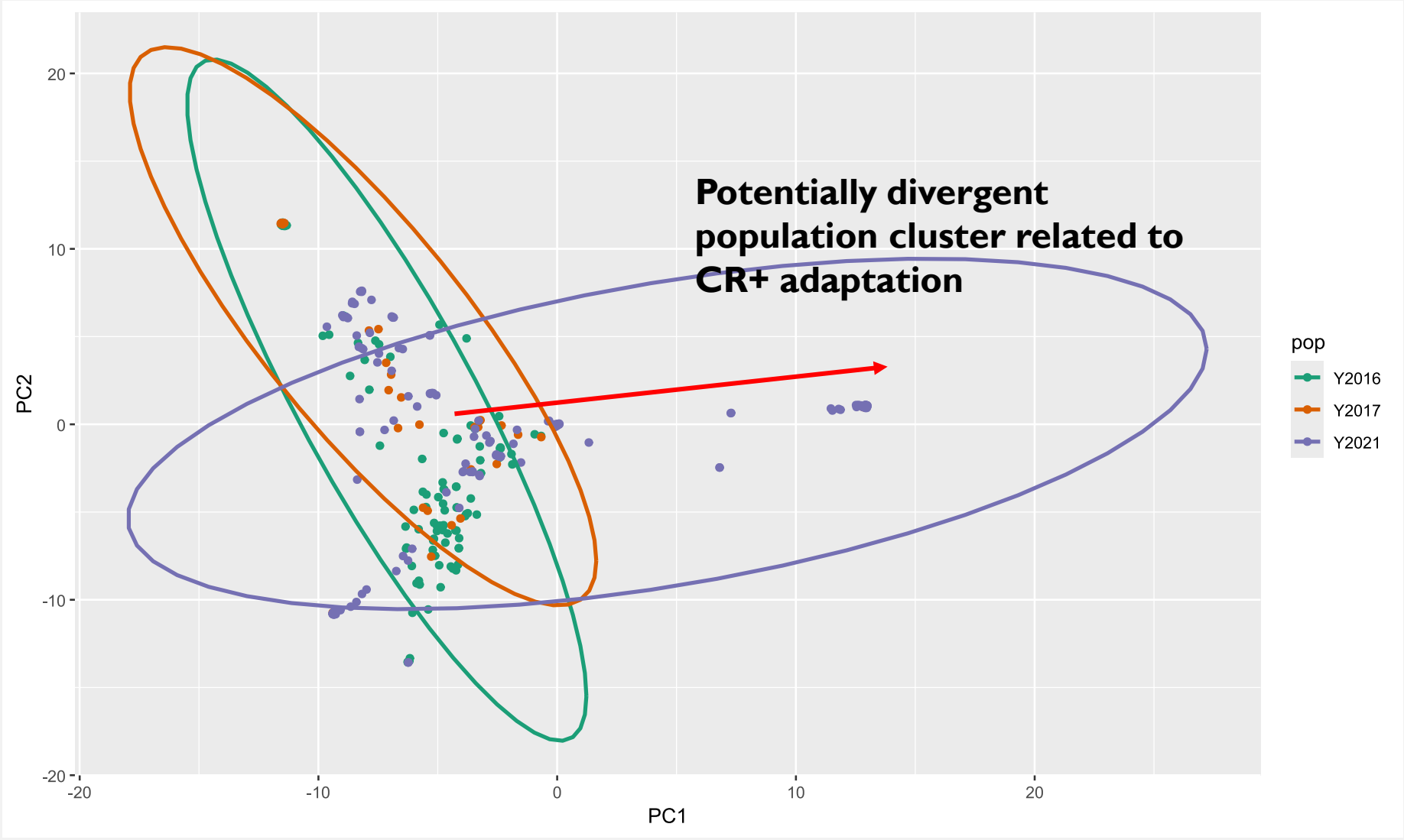
C. beticola population dynamics

2016 - 2017



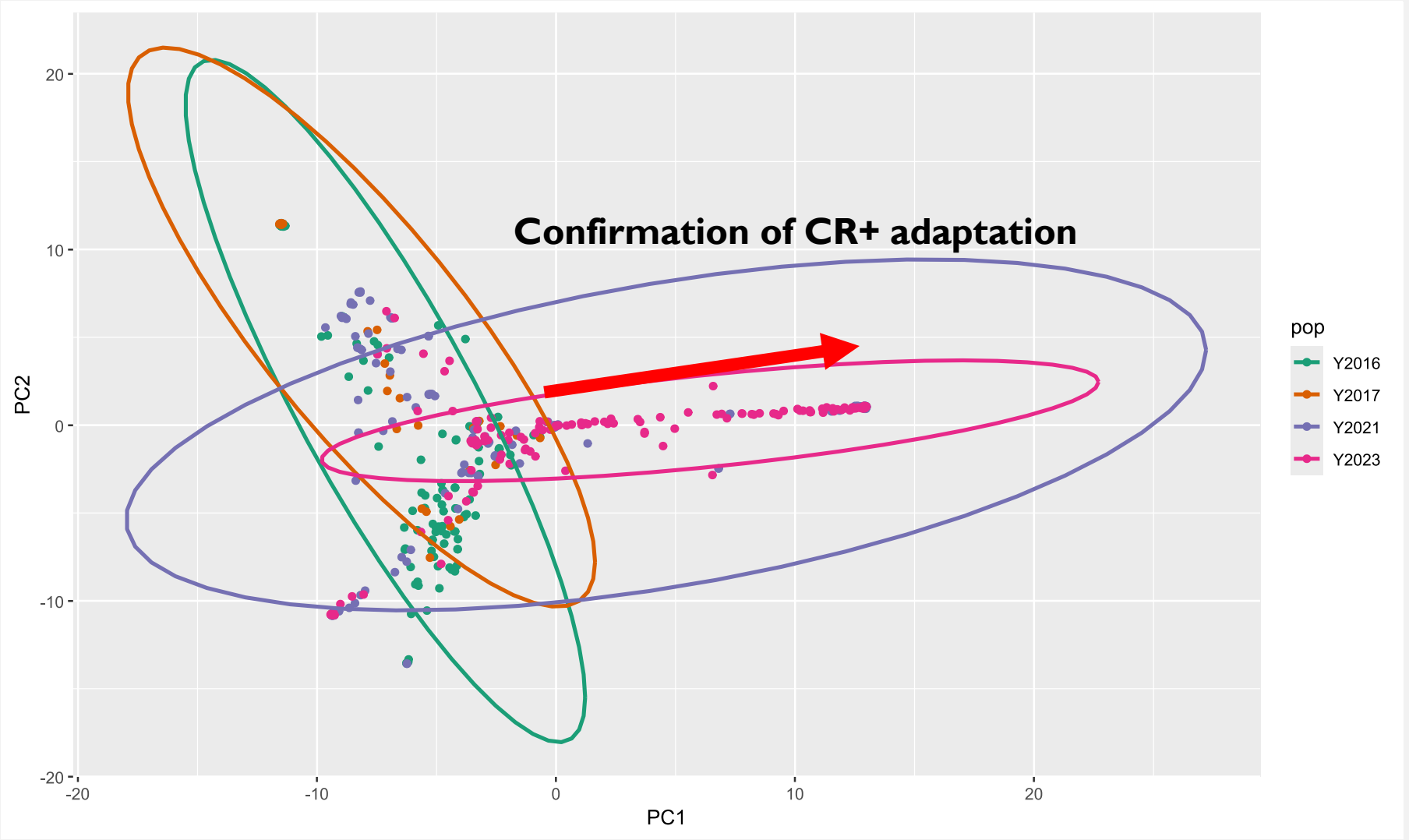
C. beticola population dynamics: CR+

2016 - 2021



C. beticola population dynamics: CR+

2016 - 2023



Ongoing reseach

- Currently finalizing an expanded CLS risk model to aid in management decisions.
- Gene knock outs of CR+ and Tin resistance genes in *C. beticola* have been successfully conducted.
- PCR based marker validation is in progress for both CR+ and Tin resistance mutations.
- Continuing *C. beticola* whole genome sequencing of isolates collected in 2024.
- Actively sequencing *C. beticola* isolates collected 1997 to present to examine population dynamics relevant to changing management practices.
  - Triazole and strobilurin introduction in early 2000's.

Acknowledgements

USDA Bolton Sugarbeet Path Lab

Melvin Bolton – Unit Research Leader

Jon Neubauer – Lab manager

NDSU Plant Pathology

Gary Secor – Group leader

Viviana Rivera-Varas - Scientist

Joe Hastings – American Crystal Sugar Company

Mark Bloomquist – Southern Minnesota Beet Sugar Cooperative

David Mettler - Southern Minnesota Beet Sugar Cooperative

Mike Metzger – Minn-Dak Farmers Cooperative

Emma Burt - Minn-Dak Farmers Cooperative

Oliver Neher – The Amalgamated Sugar Company

Research and Agriculturalist Staff who have sampled, shipped, and made this effort possible.

Sugarbeet Research & Education Board

BEET SUGAR DEVELOPMENT FOUNDATION

NDSU NORTH DAKOTA STATE UNIVERSITY

USDA U.S. DEPARTMENT OF AGRICULTURE

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Thank you for your attention!