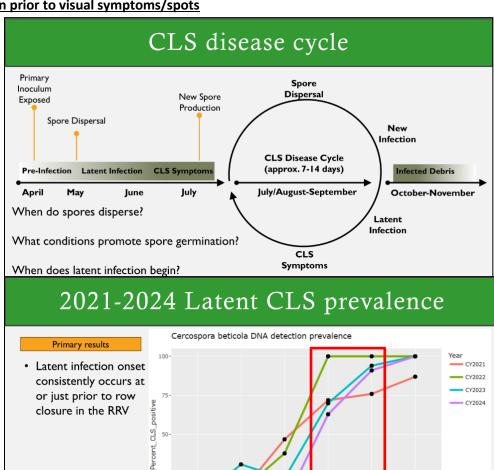
2024 Comments on Cercospora Epidemiology and Resistant Mapping

Dr. Wyatt, Dr. Bolton, Dr. Secor, and Viviana Rivera's work has shown ground-breaking advancements in understanding Cercospora Leaf Spot (CLS) epidemiology. This identified early season CLS spore dispersal from previous year's beet fields, CLS presence in leaves prior to visual detection, and that within the growing season there are dynamic resistant population shifts in response to the fungicide modes of action applied. Also, resistance for a location is not absolute in that there are susceptible isolates within a sampled population. Therefore, efficacy and CLS control can be maintained by tank mixing and rotating fungicide MOA's as well as starting early to treat primary CLS infection and delay CLS on-set in the field. Slides credited to Dr. Wyatt, Fargo USDA Epidemiologist.

Latent CLS Infection - Initial infection prior to visual symptoms/spots

Latent CLS infection period is the timeframe between the initial infection of the CLS spore in the leaf and visual detection of the Cercospora Leaf Spot. Through early season sampling, Dr. Wyatt's research has shown that by the first week of July every year virtually all locations are positive for the presence of CLS. This coincides with when the sugarbeet is at or near canopy closure. This research finding strengthens the practice of a timely initial CLS fungicide application near row closure. The initial fungicide application timing is the most critical component to get correct in the Cercospora fungicide control program and optimize revenue potential. If it is too late, the entire program is not as effective and put in jeopardy as you are already "behind".



June_4th

We

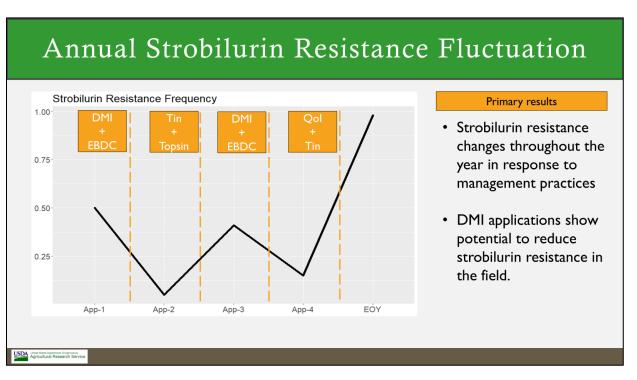
July 2nd

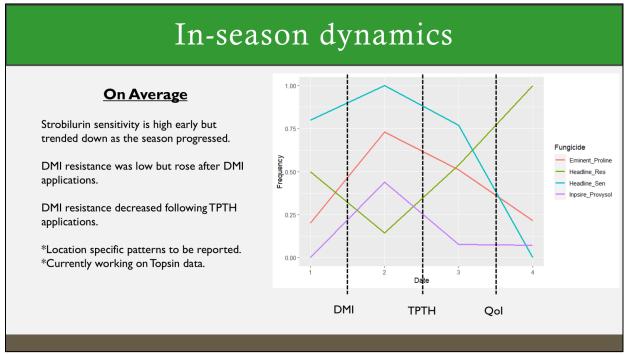
Why do we care if there are no symptoms?

ACSC Recoverable Sucrose (lbs/Acre) by first fungicide applicaiton week 2017-2023 Primary results Highest average annual RSA obtained when CLS fungicide control began in the last week of June. 2017 2018 2019 2020 2021 2022 2023 • ~5% RSA reduction from June Week 4 to July Week 1 100% 94% 92% · 2019: Low CLS pressure year 2020: High CLS pressure year · 2022: Low CLS pressure year June July July July August August Week 4 Week 1 Week 2 Week 4 Week 1 Week 2

In-Season CLS Resistant Population Shifts In Response to Fungicide Modes of Action Applied

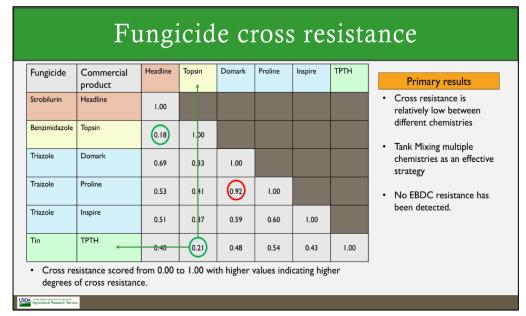
In 2022 a research program was launched with Dr. Wyatt, Fargo USDA Epidemiologist, to study how In-Season Cercospora Leaf Spot (CLS) populations shift in susceptibility/resistance levels based on what fungicide is applied. Because CLS is polycyclic in its nature (multiple generations in a growing season), resistant populations decrease or increase in response to the fungicide mode of action (MOA) that is applied. Example: If a DMI fungicide is applied, the resulting subsequent CLS population increases in resistance towards that particular DMI, but it increases the population's degree of susceptibility to the other fungicide MOA's (Ex. Tin, Headline, Topsin). Then when Tin is applied in a subsequent application it reduces the DMI resistant CLS population and raises the resistant Tin population. And so on and so forth. This interaction occurs between every fungicide application so that within the CLS season there are many susceptible CLS spores controlled by this reaction of the CLS population which allows the use of multiple MOA's throughout the Cercospora control program.





Fungicide Cross-Resistance The lower the cross-resistance factor, the greater the probability that if one fungicide mode of action won't control Cercospora, the other

fungicide mode of action won't control Cercospora, the other will. This is why we tank mix. When comparing Headline to Headline you have a fungicide cross resistance factor of "1.00" as you are comparing the likelihood of probability of cross resistance to itself. When looking at the cross-resistance factor of Tin to Topsin it is the lowest at 0.21. Another pairing



to highlight is Headline to Tin with a cross resistance of 0.40. This shows why these tank-mix partners have performed very well together. As well, there has not been any documented resistance to EBDC's making them an ideal tank-mix partner with the Triazole modes of action (Domark/Eminent, Proline, Inspire XT). **Tank-mixing and rotating these modes of action provides strength to the fungicide program limiting escapes in control & lowers resistance levels across modes of action.**

% Resistance Population Vs. % of Locations with Resistance

The traditional end-of-season samples used over the last few decades have shown that there have been increases in the % of field locations that contain fungicide resistance across all modes of action. This was measured in a way that if there was 1 out of 100 isolates from a location sample that were positive for resistance, that location was marked as positive for resistance to that fungicide to the degree it was expressed. Then this is what was shown in the resistance maps that were developed in previous years. Because of new genetic evaluation technology being

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	LI44F	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.

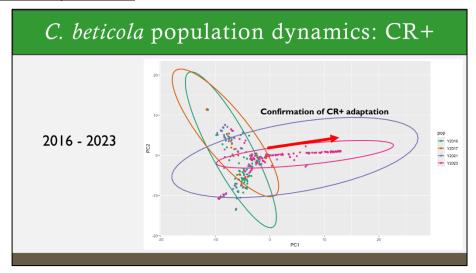
Example: Most fields sampled in 2021 had isolates that were Tin resistant.

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

used, isolates can be studied on an individual basis (quantitatively) and not as a whole general population for a location (qualitatively). Now it can be detected that even though many sample locations have isolates with fungicide resistance, the CLS isolate population demographic in those fields actually have a higher percentage of susceptibility to all fungicide modes of action than what could have previously been detected. In 2024 samples, only 25% of the Cercospora isolates were resistant to the strobilurin Headline, 15% for Topsin, 30% for Tin, 53% for Inspire/Provysol, and 62% for Domark/Eminent/Proline. This new information is groundbreaking in restoring confidence in using and tank-mixing all of our available fungicide modes of action for Cercospora control and continued resistance management.

Cercospora Population Genetic Variation - Adaption to CR+

Since before the introduction of the CR+ trait, there were naturally occurring resistant isolates to it in the environment. When the CR+ trait was introduced, it created selection pressure towards the selection and proliferation of these resistant isolates. This is expressed in the population divergence in Cercospora population mutations that became apparent in 2021 and even more confirming in 2023 (see chart). This is why it is important to



maintain a fungicide program with properly timed application intervals of 12 days to limit the propagation of these isolates. Through using proper initial application timing, tank-mixing and rotating fungicide modes of action, and high tolerance genetics (another mode of action/control) the efficacy of all tools can be optimized and maintained.

The traditional End-of-Season Cercospora Leafspot (CLS) Resistance maps

The End-of-Season Cercospora Leafspot (CLS) Resistance maps that were traditionally created represented the year end resistance levels for a location. If just one CLS isolate for that sample location was resistant, then the entire location received a resistant designation level based on that isolate. Through improved resistant measurement methods/technology it can now be detected that even though many sample locations have isolates with fungicide resistance, the CLS isolate population demographic in those fields have a higher percentage of susceptibility to all fungicide modes of action than what could have previously been detected. This was addressed in the above section: % Resistance population vs. % of Locations with Resistance. Due to being able to look at this level of granularity, the maps will not be created this year. However, to maintain an observational record of % locations/townships that had any level of a resistant population to a fungicide mode of action in the end-of-season samples the following pdf with has been created. Please click the link.

Link – Cercospora Resistant Comparison on Township 2020 – 2024 Map Grouping Table.

In 2021 – 2024 there were 17 sample locations/Agriculturist. This is based on samples that are collected at the end of the growing season, in mid-September and analyzed by Dr. Secor and Viviana Rivera-Varas at NDSU>

Starting in 2019 for the DMI fungicides, (Eminent, Inspire, Proline & Provysol) Dr. Secor's research lab provided measurements of resistance levels greater than 10ppm for EC50 values. Being able to have these observations provided greater resolution in the strength of resistance to the DMI fungicide populations. Prior to 2019 the map only showed EC50's as greater than 10ppm was preserved but a new map was created to show the addition of the detailed data of greater than 10ppm measurements and this map will be referenced in the below comments. This map was segmented for all DMI's by adding "Very High Level" categories of: 10.0001 – 25.0 ppm (VH1); 25.0001 – 50.0 ppm (VH2); >50.0 ppm (VH3). Creating these segments allows the use of the actual ppm measurements greater than 10ppm in creating the township averages.

<u>To Note:</u> Levels are based on the <u>average</u> township severity rating based on all samples taken in that township. Individual sample locations within the township may be below or above the average level of resistance for the township. As well, there are individual isolates in each sample location that are susceptible.

General Comments -

It is extremely important to have a well-timed initial fungicide application, prior to or at row closure, to keep Cercospora from becoming established in fields. Continue diligent CLS management by tank-mixing and rotating fungicide modes of action is highly recommended to aid in reducing overall CLS infections and inoculum. Using 15-20 gallons per acre water at 80+ psi, starting spray program earlier and 12-day maximum spray intervals are all essential practices that need to implemented. It is recommended that glyphosate and other herbicide applications be made separate from fungicide applications as water volumes vary by targeted pest.

For more information on CLS management, contact your Agriculturist. The American Crystal Sugar Company website https://www.crystalsugar.com/ also has detailed information. Another useful website is www.sbreb.org for the latest in North Dakota State University and University of Minnesota research information.