

EVALUATING FUNGICIDE PROGRAMS FOR CONTROL OF *CERCOSPORA* LEAF SPOT AND RELATIONSHIP TO LATENT INFECTION AND FUNGICIDE RESISTANCE PROFILES DURING THE GROWING SEASON

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INTRODUCTION

Cercospora leaf Spot (CLS) caused by fungal pathogen, *Cercospora beticola*, is the most economically significant foliar disease of sugarbeet in Minnesota and North Dakota. This disease limits sugar production by reducing both yield and sucrose concentration at harvest. Integrated approaches focusing on the use of tolerant varieties, cultural practices such as crop rotation, residue management, deep tillage, with timely application of effective fungicides can play a crucial role in limiting CLS severity and ultimately sugar and yield loss.

Fungicide applications have long been one of the most effective and significant tools in managing CLS (Khan and Smith 2005). The growers of ND and MN mostly rely on demethylation inhibitors (DMIs; FRAC 3), triphenyltin hydroxide (tin/TPTH; FRAC 30) and quinone outside inhibitors (strobilurins/QoIs; FRAC 11) to control CLS. Frequent use of fungicides with same mode of action coupled with the polycyclic life cycle and prolific sporulation of *C. beticola* are risk factors in developing fungicide resistance (van den Bosch 2014), increasing selection pressure and opportunity for mutation (Bolton et al. 2012). In populations of *C. beticola*, reduced fungicide sensitivity has been detected in several key active ingredients used for growing seasons. These include tin, thiophanate methyl or methyl benzimidazole carbamate (MBC; FRAC 1), multiple DMIs and QoIs (Secor et al. 2023). Although DMI and QoI resistant isolates may show reduced ability to infect sugarbeet in compared to sensitive isolates in absence of fungicides, this reduction is minimal but enough for fungicide-resistant isolates to persist over-time (Liu et al. 2023). Therefore, practices have shifted from individual treatments to tank mixing fungicides and mode of action rotation in programs (Khan and Nelson 2005) limiting the growth of fungicide-resistant isolates of *C. beticola*. Evaluation and selection of fungicides, initial spray timing, and application sequences has been a priority of growers and cooperatives of ND and MN to control CLS.

This project focuses on assessing the performance of spray programs with early first applications, relationship between different fungicide programs, latent *C. beticola* infections, presence of fungicide-resistant isolates, and yield parameters and quality at harvest.

Objectives of the project:

- Assess the performance of different fungicide programs to control Cercospora leaf spot on CR+ and non-CR+ varieties and effect on yield and quality at harvest
- Evaluate the relationship between latent *C. beticola* infections and pre-symptomatic fungicide applications
- Investigate changes in resistance profiles of *C. beticola* populations following fungicide applications throughout the growing season.

METHODS and MATERIALS

Field Trials

The experiment was conducted at two locations near Foxhome, MN and near Kragnes, MN. At each location, two similar trials were conducted, one planted with a CR+ variety, Beta 7231 that had a 2-year-average 2.0 CLS rating and a non-CR+ variety, Crystal 912 that had a 2-year average 5.0 CLS rating (Brantner and Moomjian 2023). Standard seed treatments were used. Planting dates were April 25th at Foxhome, and May 9th at Kragnes. Each plot consisted of six 30-foot-long rows with 22-inch spaces in randomized complete block design with four replications. The data of CLS severity rating was collected throughout the season. The scale developed by Jones and Windels

(1991) was used to rate disease severity in the center two rows of each plot. Area under disease progress curve (AUDPC) was calculated from CLS severity and used to compare disease severity between plots across both locations.

Plots were inoculated with *C. beticola* infested leaf material from 2024 season, mixed with talc in 3:2 ratio, at a rate of 5.0 lbs per acre. Inoculations were performed at Foxhome on July 9th and Kragnes on July 10th. Only the four center rows (2-5) were treated using a tractor-mounted CO₂-pressurized boom sprayer calibrated to 17 gallons per acre at 60 psi. The treatments varied by date of the first fungicide application date and interval between subsequent applications (Table 1). Fungicide sequence and materials used is indicated in Table 2. Yield and recoverable sugar were assessed at harvest on September 22nd at Foxhome and on September 24th at Kragnes. At harvest, plots were first mechanically defoliated and then the center two rows were harvested. Randomly selected roots, weighting approximately 25, were sent to the American Crystal Sugar Company Quality Tare Laboratory, East Grand Forks, MN and analyzed for sugar quality. The effect of treatments on yield, recoverable sugar, and area under the disease progress curve (AUDPC) was evaluated in R version 4.5.0.

Table 1. Treatment list and application schedule description of treatments for the sugarbeet field trials conducted near Kragnes, MN, and Foxhome, MN in 2025. One CR+ and one CLS-susceptible variety were used at each location.

Treatment	Time of first Application	Application Interval	Number of Applications	Application Dates
1	Mid-June	10-14 days	6	A B C D E F
2	Late June	10-14 days	5	B C D E F
3	Late June	Based on DIV*	4	B D E F
4	Late June	10-14 days then 21-28 days	4	B C E F [†]
5	Early July	10-14 days	4	C D E F
6	Early July	10-14 days then based on DIV	4	C D E F
7	Early July	10-14 days then 21-28 days	4	C D F
8	Disease Onset	10-14 days the based on DIV	3	D E F
9	3-5% CLS Severity	10-14 days	3	D E F
10	Non-treated Check	NA	0	NA

*DIV= Daily Infection Value

**Application dates at Foxhome: A = June 19th, B = July 1st, C = July 14th, D = July 30th, E = August 15th, F = August 25th., Application dates at Kragnes: A = June 19th, B = July 2nd, C = July 17th, D = July 31st, E = August 12th, F = August 27th.

[†] Treatment 3 had a shorter final spray interval than originally intended.

Table 2. The same fungicide modes of action and tank mix partners were used for all treatments each trial conducted in 2025. Treatments with later fungicide program start dates did not use all six applications.

Application	Mode(s) of action	Product @ Rate
1st	EBDC	Koverall @ 2 lbs/A
2nd	DMI (Tetraconazole) + EBDC	Minerva @ 13 fl oz/A + Koverall @ 2 lbs/A
3rd	Tin + EBDC	Super Tin @ 8 fl oz/A + Koverall @ 2 lbs/A
4th	DMI (Difenoconazole, Propiconazole) + EBDC	Inspire XT @ 7 fl oz/A + Koverall @ 2 lbs/A
5th	Tin + EBDC	Super Tin @ 8 fl oz/A + Koverall @ 2 lbs/A
6th	Copper + EBDC	Badge SC @ 2 pt/A + Koverall @ 2 lbs/A

Molecular assays for CLS detection and fungicide resistance profiling

Sugarbeet leaf samples were collected from each of the center two rows prior to each fungicide application throughout the growing season. Each sample consisted of three leaves taken at approximately 5-foot intervals from within the rows. Following each leaf sampling event, *Cercospora beticola* DNA was extracted and processed by the Wyatt lab at the USDA-ARS Sugarbeet Unit in Fargo, ND per previously described protocols (Wyatt 2024). Briefly, 10 leaf disks were hole-punched from each leaf sample and freeze-dried. Following DNA extraction, sample DNA was subjected to qPCR assays to detect QoI fungicide resistance (G143A mutation) (Bolton et al. 2013), DMI fungicide resistance (E170 and L144F mutations) (Spanner et al. 2021; Shrestha et al. 2022), and benzimidazole (MBC) fungicide resistance (E198A). DNA extractions were conducted within 24 hours of leaf sample collection and stored frozen until qPCR was completed.

RESULTS AND DISCUSSION

The cumulative rainfall during the growing season was 33.7 and 18.7 inches from the date of planting to date of harvest at the Foxhome and Kragnes locations, respectively. At the Foxhome site, the average CLS rating in the nontreated control plots was 7.8 at harvest, while the equivalent plots at the Kragnes site had a rating of 5.4. This variation in CLS disease pressure was likely influenced mainly by environmental factors. At both locations, the average daily temperature high was 67°F from June 1st through harvest. Rainfall had a greater influence on disease risk. There were relatively high DIVs recorded in 2025 by the North Dakota Agricultural Weather Network (NDAWN) at Foxhome compared to Kragnes (Glyndon weather station). NDAWN recorded cumulative DIVs of 188 at Foxhome and 159 near the Kragnes site. As a result, there was no difference between the fungicide application schedules of treatments 4 and 5, and between treatments 8 and 9 when DIV was considered in these particular trials. Due to the need to avoid wet conditions and waterlogged soil, late June applications were actually made during the first few days in July. This delay reflects grower practices in the region.

At the Kragnes location, for both CR+ and non-CR+ varieties, the fungicide program that began in mid-June and late June, six applications or five applications respectively, with subsequent applications made at approximately 14 days intervals (“standard intervals”) had significantly lower CLS disease severity compared to non-treated control plots (Table 3). Similarly, at Foxhome location, regardless of sugarbeet varietal tolerance to CLS, fungicide programs that began in mid-June with standard application intervals had the lowest AUDPC value, indicating reduced CLS severity. For CR+ sugarbeet, fungicide programs initiated either in late June, early July, 3-5% severity or disease onset was reported having statistically similar level of CLS control ($P = 0.002$). However, on non-CR+ sugarbeet, spray programs that began in late June or early July (with standard interval or extended interval) significantly improved disease control compared to programs starting at 3-5% severity or disease onset. These treatments only had three fungicide applications during the season, and had significantly increased CLS severity ($P = 0.03$), similar to non-treated control plots.

As observed in these experiments, for both CR+ and non-CR+ varieties, early fungicide applications were associated with increased recoverable sugar per acre (RSA), but not always at a statistically significant level. In Kragnes location, the spray program initiated on early July and late June had higher RSA for non-CR+ variety ($P = 0.001$) and CR+ variety ($P = 0.05$) respectively. Similarly, at the Foxhome location, late June and mid-June program start dates resulted in significantly higher recoverable sugar for non-CR+ ($P = 0.04$) and CR+ ($P = 0.001$) varieties, in comparison to later programs start dates. Decreased RSA was reported in non-treated control as well as in the spray program that began at disease onset in both the locations, despite differences in CLS pressure.

Complete analysis of fungicide resistance profiles from the 2025 season is underway and will be prepared for publication and distribution in the *Journal of Sugar Beet Research* or comparable academic journal in combination with results from the 2024 season.

Table 3. Effect of fungicide program start date and interval on Cercospora leaf spot disease severity (area under the disease progress curve), yield, and recoverable sugar per acre (RSA) in CR+ and non-CR+ sugarbeet at field trial near Foxhome, MN and Kragnes, MN in 2025.

Location/ Variety	Program start date/ Intervals ¹	CLS Severity (AUDPC) ²	Yield (tons/A)	RSA ³ (1000 lbs)
Foxhome/ CR+	Mid-June/ Standard	71.2 a	34.5 a	14.2 a
	Late June/ Standard	83.3 ab	31.7 a	13.9 ab
	Late June/ DIV	82 ab	33.1 a	14 ab
	Late June/Extended	74.6 a	27.2 ab	13.7 abc
	Early July/ Standard	93.8 ab	30.4 ab	13.8 ab
	Early July/ DIV	96.3 ab	27 ab	13.6 abcd
	Early July/ Extended	100.9 abc	26.9 ab	13.4 bcd
	Disease Onset/ DIV	141.5 cd	26.2 ab	12.9 de
	3-5% CLS Severity/ Standard	123.1 bc	26.5 ab	13.1 cde
	Non-treated Check	176.1 d	20.3 b	12.4 e
<i>P</i> =	0.002	0.001	NS ⁴	
Foxhome/ non-CR+	Mid-June/ Standard	90.6 a	40.2 a	12.5 a
	Late June/ Standard	109.5 ab	33.4 abc	12 ab
	Late June/ DIV	106.2 ab	32.1 abc	12.6 a
	Late June/Extended	108.7 ab	37.5 ab	12.1 ab
	Early July/ Standard	147.2 bc	29.9 bc	11.4 bc
	Early July/ DIV	146.8 bc	32.8 abc	11.3 bc
	Early July/ Extended	164.9 c	25.9 c	10.7 cd
	Disease Onset/ DIV	224.6 d	28.5 bc	10.6 cd
	3-5% CLS Severity/ Standard	224.1 d	27.4 c	10 d
	Non-treated Check	255.6 d	28.7 bc	9.7 d
<i>P</i> =	0.03	0.04	0.04	
Kragnes/ CR+	Mid-June/ Standard	11.4 a	35.1 ab	14.9 ab
	Late June/ Standard	13.2 ab	37.3 a	15.6 a
	Late June/ DIV	16.9 ab	33.1 ab	15.2 ab
	Late June/Extended	13.8 ab	33.2 ab	15 ab
	Early July/ Standard	23.6 bcde	34 ab	14.8 ab
	Early July/ DIV	25.2 cde	32.1 ab	15.1 ab
	Early July/ Extended	21.2 abc	34.2 ab	14.7 ab
	Disease Onset/ DIV	30.9 de	33 ab	14.7 ab
	3-5% CLS Severity/ Standard	35.1 d	29.3 ab	15.4 a
	Non-treated Check	68.1 e	26.2 b	14.2 b
<i>P</i> =	<0.001	0.05	NS	
Kragnes/ non-CR+	Mid-June/ Standard	41.1 a	37.3 ab	12.9 ab
	Late June/ Standard	51 ab	37.7 ab	12.1 ab
	Late June/ DIV	66.9 abcd	39.8 a	13.1 a
	Late June/Extended	59.1 abc	38.2 ab	13 a
	Early July/ Standard	89.9 cdef	33.5 ab	12.6 ab
	Early July/ DIV	85.8 bcde	35.2 ab	13 a
	Early July/ Extended	65.6 abcd	38.4 ab	13.1 a
	Disease Onset/ DIV	101 def	32.4 ab	11.9 ab
	3-5% CLS Severity/ Standard	110 ef	36.1 ab	11.5 bc
	Non-treated Check	126.8 f	28.6 b	10.4 c
<i>P</i> =	< 0.01	0.001	0.05	

¹Standard = 10-14 days; Extended = 10-14 days, then 21-28 days; DIV = applications made as indicated by Daily Infection Value

²Area Under the Disease Progress Curve

³Recoverable Sugar per Acre

⁴Non-significant

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