

EVALUATION OF FUNGICIDE SPRAY PROGRAMS TO MANAGE CERCOSPORA LEAF SPOT USING CR+ AND NON-CR+ SUGARBEET VARIETIES, 2023

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INTRODUCTION

Cercospora leaf spot (CLS), caused by the fungus *Cercospora beticola*, continues to be a challenge to sugarbeet growers in Minnesota and North Dakota, especially when growing conditions are warm and humid. The management of CLS must incorporate integrated practices such as conventional tillage, crop rotation, and spatial separation from previous sugarbeet fields when possible. Variety selection is also a critical aspect in managing CLS, but each variety has unique characteristics regarding yield, sugar quality, and disease tolerance. Additionally, the use of effective fungicides and proper timing of applications can significantly delay CLS development and reduce the extent of economic losses. However, with the increasing incidence of fungicide-resistance in *C. beticola* isolates across sugarbeet growing regions of Minnesota and North Dakota (Secor et al. 2023), the use of highly tolerant sugarbeet varieties (i.e., CR+ varieties) may be vital in managing CLS disease (Mettler and Bloomquist 2021, 2022).

In 2021, conidia of *C. beticola* had been identified in spore traps as early as May 03 in some growing regions (Secor et al. 2022). Several weeks before leaf spot symptoms were visible, the DNA of *C. beticola* was also detected in sugarbeet leaves in early June of 2020 (Bloomquist et al. 2021) and June of 2021 (Secor et al. 2022). Once the detached conidia land on the sugarbeet leaf or petiole, the fungus can initiate infection under favorable environmental conditions. Results from Rivera-Varas (2021) indicate that conidia can germinate within 2 hours even at 10°C; however, optimal temperatures for germination and infection are 25-35°C (Jacobson and Franc 2009). Following infection, leaf spot symptoms can develop within 5 days (Solel and Minz 1971), and secondary conidia can form after 7 days under favorable conditions (Jacobson and Franc 2009). The development of CLS symptoms and secondary conidia are highly influenced by temperature, humidity, light, leaf age, and disease tolerance of the host. Generally, infection cycles are prolonged as CLS tolerance of the host increases (Jacobson and Franc 2009). Although pathogen growth is not completely stopped in CLS-tolerant varieties, the plants have less ROS production and express lower levels of disease severity compared to CLS-susceptible varieties (Bhuiyan et al., 2023). Bhuiyan et al. (2021, 2023) also reported that infection of *C. beticola* and the hypersensitive response of the host is delayed in a CLS-tolerant variety, implying that the development of secondary conidia is also delayed. In field conditions, Bhandari et al. (2023) reported that the first CLS symptoms were observed on CLS-susceptible varieties 13 days prior to CLS-tolerant varieties. Metzger (2021) reported that the final CLS disease severity of CR+ varieties is significantly less compared to susceptible varieties in the 2020 Minn-Dak Farmers Cooperative (MDFC) CLS Nursery near Foxhome, MN. Two trials in separate locations were conducted in 2020 and 2021 by the Southern Minnesota Beet Sugar Cooperative (SMBSC) to determine the best fungicide program to pair with varieties with differing levels of CLS tolerance. Mettler and Bloomquist (2021, 2022, 2023) report from the field trials that highly tolerant varieties do not need the same rigorous fungicide program that moderately susceptible varieties need to produce good yields. Lien et al. (2023) also reported that disease pressure on a CLS-tolerant variety was very low, and yields were similar regardless of the fungicide spray programs, ranging from 1 to 6 applications, in a field trial conducted in Crookston, MN. Since 2021, CR+ sugarbeet varieties with traits that impart improved tolerance to CLS are now available to growers throughout Minnesota and North Dakota. Promisingly, these newly released varieties are coupled with improved performance and can produce a recoverable sucrose per acre that is comparable to susceptible varieties. Additionally, it is hoped that the cost of fungicide management can be reduced by integrating these varieties and decreasing the number of fungicide applications.

OBJECTIVES

The trial objective is to evaluate a CR+ variety and standard fungicide programs with different timings for 1) the relative control of CLS disease on sugarbeet, and 2) the effect on harvestable root yield and sucrose quality.

MATERIALS AND METHODS

The trial was established as a randomized complete block design with 4 replicates at the University of Minnesota Northwest Research and Outreach Center in Crookston, MN. Field plots were fertilized for optimal yield and quality. A moderately susceptible (MS) variety (Crystal 912RR) with a 2-year average *Cercospora* rating of 5.0 (Brantner and Moomjian 2023) and a CR+ variety (Crystal 021RR) with a 2-year average *Cercospora* rating of 2.2 (Brantner and Moomjian 2023) was used. All seed was treated with standard seed treatments and were sown in 6-row by 35-foot long plots at 4.5-inch spacing in 22-inch rows on May 10. Plant stands were evaluated June 01 by counting the number of plants in the center two rows of each plot to verify an average plant population of 192 plants per 100 ft of row for Crystal 021 and 224 plants per 100 ft of row for Crystal 912.

On June 27 (14 to 16-leaf stage), all rows within each plot were inoculated with a mixture of fine talc and dried ground CLS-infected sugar beet leaves (1:2 weight by weight) using a Nalgene® 1L bottle to deliver a rate of 4.5 lbs. per acre (3 grams of mixture per 35 feet of row). CLS-infected sugar beet leaves used for the inoculum were collected from nontreated rows at the end of 2022 growing season. Fungicide treatments (see tables) were applied to the center four rows using a tractor-mounted 3-point sprayer with XR TeeJet 11002 VS flat fan nozzles calibrated to deliver 17.1 gallons water/A at 100 psi. Fungicides were applied approximately every 12 days depending on weather conditions. Fungicide applications began when weather was conducive for disease development and coincided with canopy closure, except for the first applications on June 23, 4 days prior to inoculation. Fungicide treatments were applied on June 23, June 30, July 12, July 24, Aug 07, and Aug 23. CLS disease severity was evaluated beginning July 11 and continued through Sept 18 using the following scale based on infected leaf area: 1=0.1% (1-5 spots/leaf), 2=0.35% (6-12 spots/leaf), 3=0.75% (13-25 spots/leaf), 4=1.5% (26-50 spots/leaf), 5=2.5% (51-75 spots/leaf), 6=3%, 7=6%, 8=12% 9=25%, 10=50%; rating scale is outlined by Jones and Windels (1991). CLS severity ratings were used to calculate the standardized area under disease progress stairs for statistical analysis (Simko and Piepho 2012, Simko 2021). On Sept 19, plots were defoliated, and the center two rows of each plot were harvested mechanically and weighed for root yield. Twelve representative roots from each plot were analyzed for sugar quality at the American Crystal Sugar Company Quality Tare Laboratory, East Grand Forks, MN. Statistical analysis was conducted in R (v 4.3.1, R Core Team 2023). A mixed-model analysis of variance was performed using the package *lmerTest* (v 3.1-3), with treatment defined as the fixed factor and replication as the random factor. Means were separated at the 0.05 significance level using the package *emmeans* (v 1.8.7) adjusted for Tukey's honest significant difference (HSD).

RESULTS AND DISCUSSION

In 2023, the Northwest Research and Outreach Center (NWROC), Crookston, MN, recorded a total rainfall of 1.71 in. for April, similar to the 30-year average of 1.32 in. However, 0.87 in. of rainfall was received in May, much less than the 30-year average of 2.81 in. The rest of the growing season was slightly drier than the 30-year average, receiving only 4.7 in. of total rainfall in June, July, and August, less than half of the 30-year average of 9.6 in. May and June were 7.6°F and 6.7°F warmer, respectively, whereas July was 4.1°F cooler compared to the 30-year averages.

Following inoculation, daily infection values monitored by the Eldred NDAWN station had risen to a moderate level; the prolonged warm temperatures and high humidity provided conditions that favored the establishment of the *Cercospora* inoculum. Despite less-than-average rainfall, disease pressure rapidly increased during the month of August. Standard fungicide programs significantly reduced total CLS severity (sAUDPS), especially the MS variety (Table 2). Disease pressure progressed in the MS nontreated control to a level above the known economic threshold of 3% severity (equivalent to a rating of 6.0) by Aug 16 and reached a rating of 9.8 by Sept 08 with complete defoliation and emergence of new foliage (Fig. 2). Fungicide programs in the MS variety that received only 3 applications beginning July 24 reached disease levels above the economic threshold by 25 Aug; whereas, the fungicide programs with 4, 5, or 6 applications remained below the economic threshold (Table 1). Disease pressure became apparent in the nontreated CR+ variety by the end of August, reaching a rating of 2.2 by Sept 08 (Fig. 2). Overall, disease pressure in the CR+ variety was minimal throughout the season, and CLS severity was very low, regardless of the fungicide spray program (Fig. 1); however, numerical differences were present in which the nontreated control and the treatment with only 1 application in the CR+ variety had a higher CLS rating than the

treatment with 6 fungicide applications in the MS variety (Fig. 2) and also resulted in a higher sAUDPS compared to treatments with 2 or more fungicide applications in the CR+ variety (Table 2). There were no significant differences in percent sugar, sugar loss to molasses (SLM), root yield, or recoverable sucrose per acre; though, numerical differences show that the nontreated control in the MS variety resulted in the lowest recoverable sucrose (Table 2).

Table 1. Select Cercospora leaf spot (CLS) 0-10 ratings associated with fungicide spray programs to manage CLS of sugarbeets in a CLS-inoculated field trial planted on May 10, 2023 at the University of Minnesota, Northwest Research and Outreach Center, Crookston.

Variety and Program ^z	Treatment(s) and timing ^y	CLS ratings (0-10)					
		Jul 11	Jul 20	Aug 03	Aug 16	Aug 25	Sept 08
CR+ 6-Spray	Provysol A + Manzate Pro-Stick ABDE + Super Tin CF + Topsin 4.5 FL C + Proline 480 SC D + Priaxor F	0.00	0.05	0.05	0.45	0.55	1.05
CR+ 6-Spray (Skip 3 & 5)	Provysol A + Manzate Pro-Stick AD + Super Tin BF + Topsin 4.5 FL B + Proline 480 SC D + Priaxor F	0.05	0.25	0.00	0.45	0.75	0.95
CR+ 6-Spray (Skip 2, 4, & 5)	Provysol A + Manzate Pro-Stick A + Super Tin C + Topsin 4.5 FL C + Proline 480 SC F + Priaxor F	0.05	0.15	0.05	0.70	0.85	1.15
CR+ 5-Spray	Provysol B + Manzate Pro-Stick BDE + Super Tin CF + Topsin 4.5 FL C + Proline 480 SC D + Priaxor F	0.10	0.20	0.00	0.70	0.80	0.85
CR+ 5-Spray (Skip 3)	Provysol B + Manzate Pro-Stick BE + Super Tin CF + Topsin 4.5 FL C + Proline 480 SC E + Priaxor F	0.05	0.15	0.00	0.60	0.90	1.00
CR+ 5-Spray (Skip 3 & 4)	Provysol B + Manzate Pro-Stick B + Super Tin C + Topsin 4.5 FL C + Proline 480 SC F + Priaxor F	0.05	0.10	0.05	0.65	0.85	1.55
CR+ 4-Spray	Provysol C + Manzate Pro-Stick CE + Super Tin DF + Topsin 4.5 FL D + Proline 480 SC E + Priaxor F	0.05	0.00	0.05	1.05	1.55	1.00
CR+ 4-Spray (Skip 3)	Provysol C + Manzate Pro-Stick C + Super Tin D + Topsin 4.5 FL D + Proline 480 SC F + Priaxor F	0.00	0.10	0.00	0.90	0.85	1.50
CR+ 4-Spray (Skip 2 & 3)	Proline 480 SC C + Manzate Pro-Stick C + Super Tin F + Priaxor F	0.10	0.00	0.00	0.90	0.85	1.25
CR+ 3-Spray (Skip 2)	Proline 480 SC D + Manzate Pro-Stick D + Super Tin F + Priaxor F	0.15	0.05	0.00	0.45	0.80	1.15
CR+ 2-Spray	Proline 480 SC E + Manzate Pro-Stick E + Super Tin F + Priaxor F	0.00	0.05	0.10	0.80	0.85	1.25
CR+ 3 Spray	Provysol D + Manzate Pro-Stick D + Super Tin E + Topsin 4.5 FL E + Proline 480 SC F + Priaxor F	0.05	0.00	0.00	0.95	1.15	1.30
CR+ 1-Spray	Proline 480 SC F + Priaxor F	0.05	0.05	0.00	1.05	1.55	2.00
CR+ Nontreated	Nontreated Control	0.00	0.00	0.25	0.90	1.70	2.20
MS 6-Spray	Provysol A + Manzate Pro-Stick ABDE + Super Tin CF + Topsin 4.5 FL C + Proline 480 SC D + Priaxor F	0.15	1.00	0.20	1.00	1.60	1.65
MS 5-Spray	Provysol B + Manzate Pro-Stick BDE + Super Tin CF + Topsin 4.5 FL C + Proline 480 SC D + Priaxor F	0.10	0.60	0.30	1.35	2.65	2.45
MS 4-Spray	Provysol C + Manzate Pro-Stick CE + Super Tin DF + Topsin 4.5 FL D + Proline 480 SC E + Priaxor F	0.35	0.95	0.65	4.00	6.15	4.90
MS 3-Spray (ACSC 1)	Provysol D + Manzate Pro-Stick D + Super Tin E + Topsin 4.5 FL E + Proline 480 SC F + Priaxor F	0.50	1.25	2.15	5.95	7.80	7.65
MS 3-Spray (ACSC 2)	Super Tin DF + Topsin 4.5 FL D + Proline 480 SC E + Manzate Pro-Stick E + Priaxor F	0.35	1.20	2.25	5.70	7.25	7.00
MS Nontreated	Nontreated Control	0.45	1.10	2.40	7.10	9.35	9.75
	<i>P</i> -value	***	***	***	***	***	***

^z Crystal 021RR with two-year Cercospora rating of 2.2 (CR+) and Crystal 912 with two-year Cercospora rating of 5.0 (MS)

^y Treatment rates per acre are as follows: Provysol = 5 fl oz, Manzate Pro-Stick = 2 lb, Super Tin = 8 fl oz, Topsin 4.5 FL = 10 fl oz, Proline 480 SC = 5.7 fl oz, Priaxor = 6.7 fl oz; Non-ionic surfactant (NIS; Permeate) was used at a rate of 0.125% v/v with Provysol and Proline 480 SC; letters represent the following dates: **A**= Jun 23, **B**= Jun 30, **C**= Jul 12, **D**= Jul 24, **E**= Aug 07, **F**= Aug 23

^x Significance codes: 0.0001 (***), 0.001 (**), 0.01 (*)

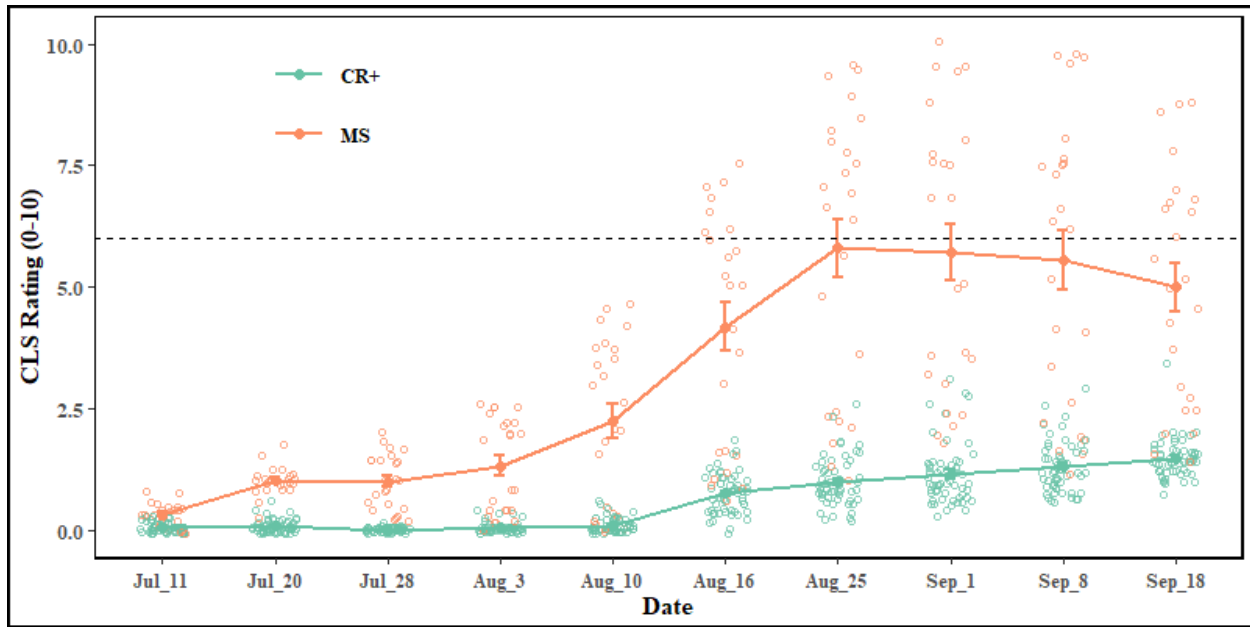


Figure 1. Cercospora leaf spot (CLS) disease severity ratings throughout the 2023 growing season for sugarbeet varieties sown on May 10 in a field trial inoculated with CLS-infested leaves on June 27 at the University of Minnesota, NWROC, Crookston, MN. Hollow dots represent each data point; filled dots represent treatment means and error bars represent the standard error of each variety. The dashed horizontal line represents the known CLS economic threshold of 3% severity, equivalent to a rating of 6.0.

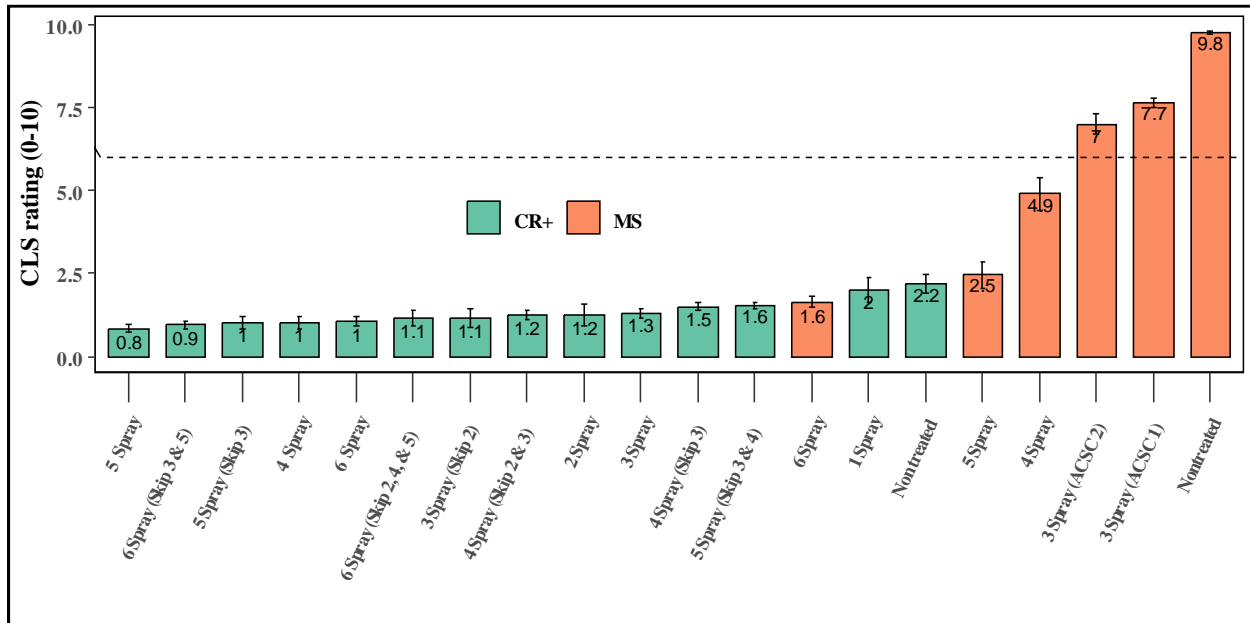


Figure 2. Cercospora leaf spot (CLS) disease severity ratings on Sept 8, 2023, for fungicide spray programs in sugarbeet varieties sown on May 10 in a field trial inoculated with CLS-infested leaves on June 27 at the University of Minnesota, NWROC, Crookston, MN. Columns display the mean for each treatment; error bars represent the standard error of each treatment. The dashed horizontal line represents the known CLS economic threshold of 3% severity, equivalent to a rating of 6.0.

Table 2. Effects of fungicide spray programs on CLS disease, harvestable yield, and sucrose quality of sugarbeets in a CLS-infested field trial planted on May 10, 2023 at the University of Minnesota, Northwest Research and Outreach Center, Crookston.

Variety and Program ^z	CLS Severity (sAUDPS) ^{y,x}	Sugar (%)	SLM (%)	Yield (tons/A)	Sucrose (lb/A)	Gross Rev. (\$/ton) ^w	Gross Rev. (\$/A) ^w	Fung. Cost (\$/A) ^v	Net Rev. (\$/A) ^w
CR+ 6-Spray	0.4 a	18.6	0.8	27.8	9904	95.68	2659.50	115.42	2544
CR+ 6-Spray (Skip 3 & 5)	0.5 a	19.4	0.9	24.7	9148	102.47	2529.78	101.82	2428
CR+ 6-Spray (Skip 2, 4, & 5)	0.6 a	19.1	0.8	31.9	11635	99.54	3180.00	88.46	3092
CR+ 5-Spray	0.5 a	18.7	0.8	30.2	10801	96.28	2907.77	108.62	2799
CR+ 5-Spray (Skip 3)	0.5 a	19.0	0.8	30.0	10914	99.59	2977.30	101.82	2875
CR+ 5-Spray (Skip 3 & 4)	0.7 ab	19.1	1.0	27.0	9948	100.60	2736.51	88.46	2648
CR+ 4-Spray	0.6 ab	18.3	0.9	30.3	10481	91.14	2748.71	101.82	2647
CR+ 4-Spray (Skip 3)	0.6 ab	18.9	0.9	28.4	10319	98.01	2810.29	88.46	2722
CR+ 4-Spray (Skip 2 & 3)	0.6 a	18.3	0.9	30.0	10487	92.10	2770.51	62.89	2708
CR+ 3-Spray (Skip 2)	0.6 ab	18.8	0.9	27.6	9880	96.44	2658.70	62.89	2596
CR+ 2-Spray	0.5 a	19.1	0.8	29.9	10896	99.83	2976.97	62.89	2914
CR+ 3 Spray	0.7 ab	18.9	0.8	29.0	10504	98.53	2853.50	88.46	2765
CR+ 1-Spray	0.9 bc	19.2	0.8	29.3	10782	101.41	2969.79	49.53	2920
CR+ Nontreated	1.0 cd	19.1	0.9	27.1	9880	99.52	2700.86	0.00	2701
MS 6-Spray	1.0 cd	18.7	0.8	30.4	10910	96.47	2945.28	115.42	2830
MS 5-Spray	1.4 d	19.0	0.7	29.2	10659	99.66	2912.10	108.62	2803
MS 4-Spray	2.9 e	18.0	0.8	30.5	10430	89.05	2711.01	101.82	2609
MS 3-Spray (ACSC 1)	4.5 f	18.9	0.8	28.4	10266	98.29	2790.19	88.46	2702
MS 3-Spray (ACSC 2)	4.3 f	19.0	0.7	27.2	9967	100.62	2733.80	72.89	2661
MS Nontreated	5.5 g	18.4	0.8	25.8	9081	93.44	2411.09	0.00	2411
<i>P</i> -value	<0.0001	0.5625	0.3385	0.0678	0.2974	0.6593	0.5583	NA	0.6601

^z Crystal 021RR with two-year Cercospora rating of 2.2 (CR+) and Crystal 912 with two-year Cercospora rating of 5.0 (MS); fungicides and application dates for each program are listed in Table 1.

^y Standardized Area Under Disease Progress Stairs (sAUDPS) is a mid-point combination of all CLS ratings and represents total CLS severity.

^x Means within a column followed by a common letter are not significantly different by Tukey's Honest Significant Difference test at the 0.05 level of significance

^w Revenue is based on the November 2023 American Crystal Sugar Company (ACSC) beet payment;

^v Fungicide cost is based on 2023 prices and does not include application costs.

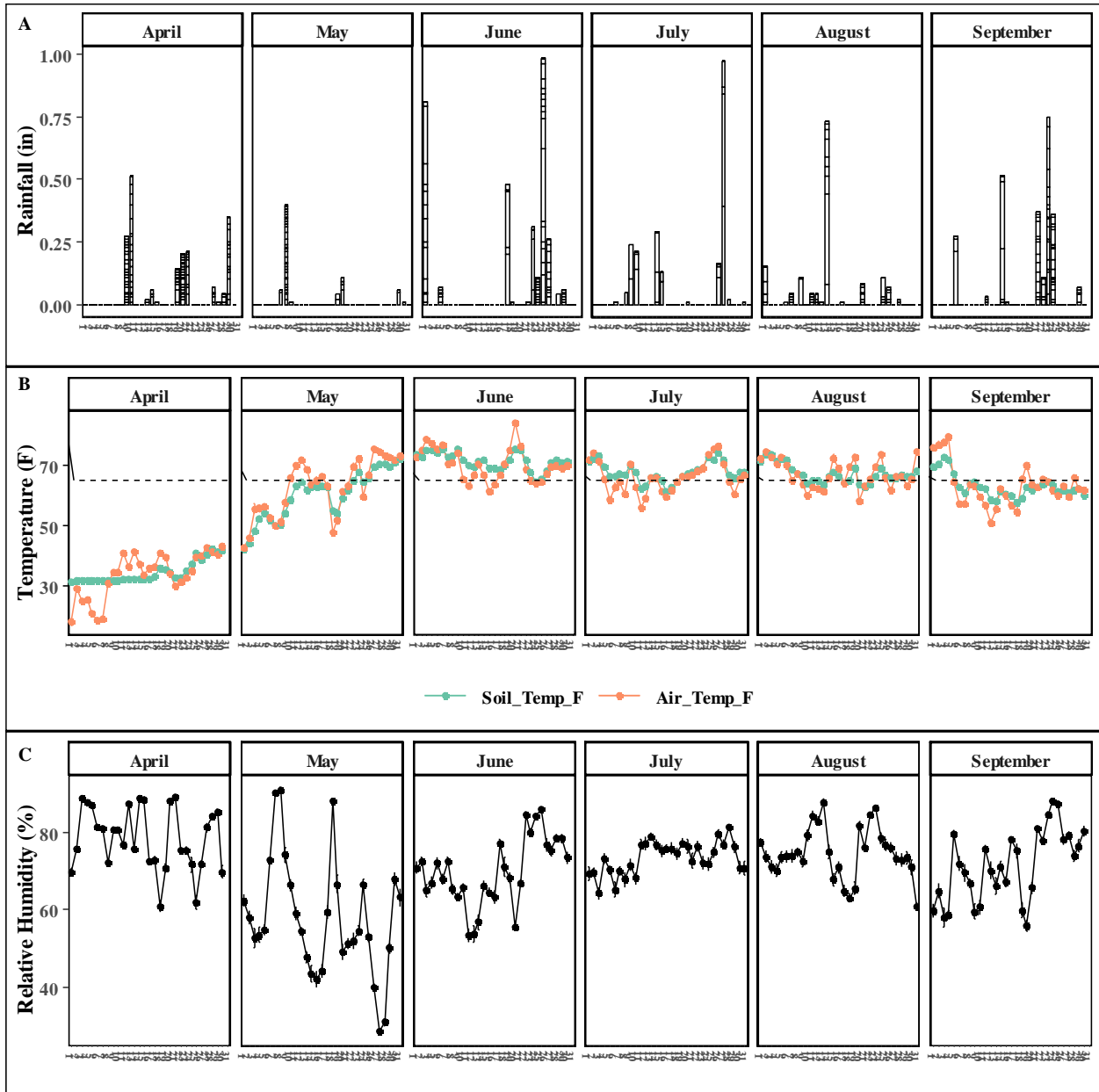
ACKNOWLEDGMENTS

We thank the Sugarbeet Research and Education Board of Minnesota and North Dakota for funding this research; BASF, UPL, and Bayer for providing chemical products; Crystal Beet Seed for providing seed; Germaines Seed Technology for treating seed; the University of Minnesota Northwest Research and Outreach Center, Crookston for providing land, equipment, and other facilities; Jeff Nielsen for plot maintenance; James Deleon, Zahra Pagiri, Stephanie Melby, and Kenan McQueen for technical assistance; American Crystal Sugar Company Quality Laboratory, East Grand Forks, MN for sugarbeet quality analysis.

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SUPPLEMENTARY FIGURES



Supplementary Fig. S1. Daily rainfall totals (A), daily mean air temperature and 4-inch soil temperature (B), and daily mean relative humidity (C) for the 2023 growing season at the NWROC weather station in Crookston, MN. The dotted horizontal line represents 65°F.