Epidemiological studies of Cercospora leaf spot of sugar beet for improved management

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Background:

Cercospora leaf spot (CLS)is the most important foliar disease of sugar beet in Michigan and several other sugar beet growing regions (Harveson et al. 2009; Lartey et al. 2010). This research aims to identify, develop, and deploy novel short-term and long-term CLS management strategies. Observations made of the disease, including early-season inoculum presence, changes in *Cercospora beticola* fungicide resistance, and performance of CLS prediction models have helped us to identify opportunities for further improvement in CLS management. Strategies which were investigated to aid in CLS management include creating improved disease prediction tools through an innovative spore abundance model, identifying alternative strategies to reduce inoculum survival for long-term management, and evaluating fungicide resistance management tactics in Michigan *C. beticola* populations. Continued population monitoring, model development and refinement, and multi-year and -location validation is ongoing.

Methods:

Objective 1. Monitor *C. beticola* spore presence and abundance using spore traps and sentinel beets to refine existing predictive modeling tools. Aerial spores were captured using Burkard spore traps and highly susceptible sentinel beet traps at the MSU Crop and Soils Farm, Frankentrost, MI, and Ontario, Canada in 2019 and the Saginaw Valley Research and Extension Center (SVREC) in 2020. Environmental factors were monitored using on-site or local MSU Enviroweather stations and evaluated for correlations to spore abundance. Stepwise regression analyses were conducted to assess the accuracy of the model variables separately and together.

Objective 2. Assess potential end-of-season management strategies to reduce inoculum levels and disease. Treatments included a nontreated control, plowing immediately post-harvest, applying heat with a burner at 1 mph prior to defoliation at-harvest, and applying a desiccant (saflufenacil, Sharpen) 7 days pre-harvest. Leaf samples were collected from each plot at harvest before topping and evaluated 0-, 45-, 90-, and 135-days post-harvest to assess *C. beticola* survival over the winter. Survival was determined by observing the percentage of lesion sporulation and viability. In 2020, highly susceptible sentinel beets and bi-weekly CLS ratings in re-planted plots were used to assess the long-term efficacy of inoculum reduction strategies. Yield and sugar data were also collected to assess the long-term efficacy of inoculum reduction strategies. A repeated trial was initiated in 2020 with the addition of a second burner treatment applied at 3 mph.

Objective 3. Determine fungicide sensitivity of *C. beticola* populations recovered from resistance management trials. Treatment programs evaluated at the SVREC included: a nontreated control; a mixed application, where both high-risk (pyraclostrobin) and low-risk (mancozeb) fungicides were applied at each spray timing; high-low, where alternate sprays of pyraclostrobin and mancozeb were applied, with pyraclostrobin sprayed first; and low-high, which is similar to the previous treatment but with low-risk applied first. Symptomatic leaves were sampled from field trials in July (after three treatments) and September (after all six treatments). Mono-conidial *C. beticola* isolates were then tested for in vitro pyraclostrobin sensitivity. A spiral gradient dilution method was used to find the effective concentration inhibiting growth by 50% (EC50). Resistance management trials were conducted in 2019 and 2020.

For all objectives, statistical analyses (analysis of variance and simple and linear mixed model regression) were conducted in SAS v. 9.4 and evaluated at the α=0.05 significance level. Fisher's protected Least Significance Difference was used for mean comparisons.

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Results & Conclusions:

Objective 1. In 2019, a preliminary model to predict spore number was developed using significantly correlated weather predictors ($R^2 = 0.23$, $P < 0.0001$). The initial model predicted daily spore abundance based on daily total precipitation, minimum daily relative humidity, maximum daily soil temperature, and maximum daily wind speed. With additional observations from Ontario in 2019 and Michigan in 2020, significant correlations were observed between spore abundance and maximum air temperature ($r = 0.35$, $P < 0.0001$) and maximum soil moisture ($r = 0.22$, $P < 0.05$), though precipitation ($r = 0.12$, $P = 0.11$) and maximum wind speed ($r = 0.17$, $P = 0.06$) were also noted. Additional locations and years will be added to the spore abundance and initial disease observations for further model refinement and validation. A preliminary model will be used in field validations conducted in 2021. Initial detections and general trends of abundance suggest a spore presence or abundance model will complement existing tools to better predict early-season risk and improve subsequent CLS management.

Objective 2. In 2019, significant treatment differences were detected in percentages of lesion sporulation ($P < 0.0001$) and lesion viability ($P < 0.05$) in at harvest samples (N=133 leaves and 240 lesions). In 2020, reduced numbers of CLS lesions were observed on sentinel beets collected in 2019 burner treated plots from May 26-June 1 ($P < 0.05$, Fig. 1A) and June 2-9 ($P < 0.01$, Fig. 1B). The heat treatment also significantly reduced the area under the disease progress curve (AUDPC), calculated from ratings in re-planted plots ($P < 0.01$, Fig. 1C). In the repeated trial initiated in 2020, lesion sporulation was reduced in at harvest ($P < 0.0001$, Fig. 2) and 45-days post-harvest samples ($P < 0.01$). Continued monitoring will occur until harvest in 2021. Novel management strategies, particularly the use of a foliar burner at-harvest, have the potential to reduce inoculum overwintering and aid in long-term CLS control.

Remaining leaf samples from inoculum overwintering studies will continue to be evaluated for the repeated trial initiated in 2020. In 2021, early-season spore presence and abundance, weekly disease ratings, and final yield and sugar data will be collected to validate the long-term efficacy of inoculum reduction strategies.

Objective 3. In 2019, no significant differences were found in mean pyraclostrobin EC_{50} values for isolates collected from the fungicide treatment programs in July (N=145 isolates) or September (N=75 isolates, *in progress*). All programs resulted in similar yields ($P < 0.001$), relative area under the disease progress curves (RAUDPC; $P < 0.01$) and performed better than the non-treated control. So far, 43% of isolates from July and 20% of isolates from September are considered highly resistant ($EC_{50} \geq 25$ ppm). All isolates tested were sensitive to pyraclostrobin concentrations below label rates (1,200-1,500 $\mu g\ ml^{-1}$). In 2019, resistance management tactics were found to have little effect on mid-season populations of *C. beticola*.

Testing of the remaining end-of-season *C. beticola* populations from 2019 and 2020 is in-progress and will continue. These samples received the full-season treatments. Isolate pyraclostrobin sensitivity will be tested and results will be evaluated by treatment. In 2021, *C. beticola* populations will be monitored for sensitivity to critical fungicide groups.

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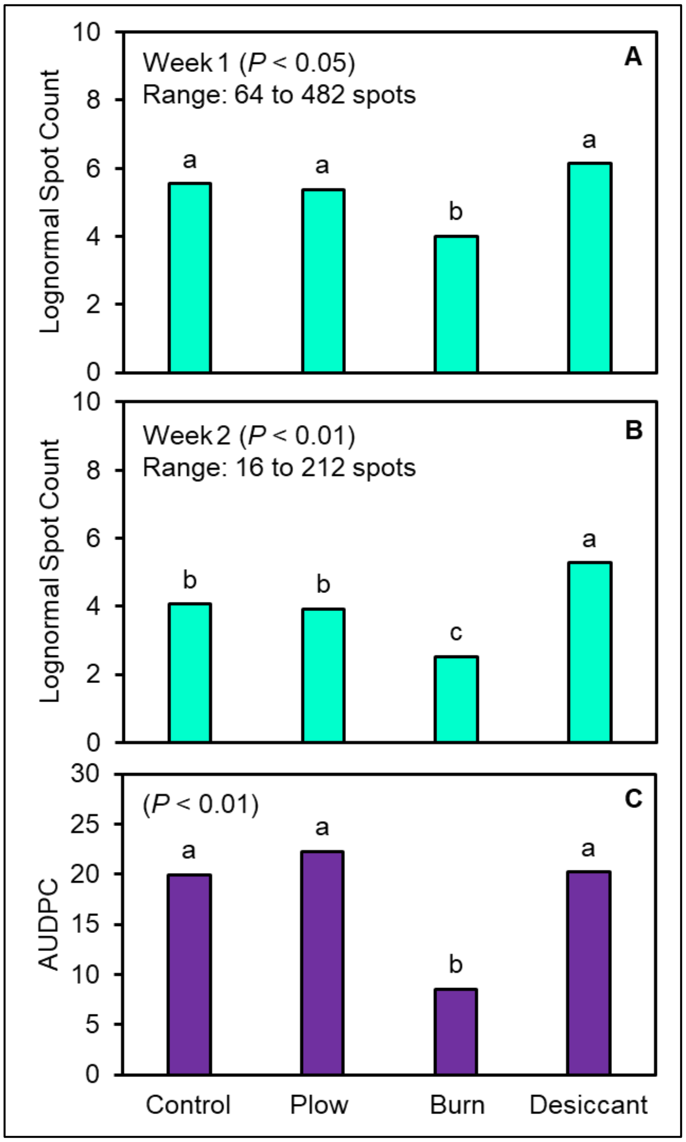

***Figure 1.** Early-season inoculum and subsequent CLS observations in 2020 following end-of-season treatments applied in 2019. Leaf spot counts were collected from sentinel beets placed in plots between **A,** May 26-June 1 and **B,** June 2-9. Subsequent bi-weekly CLS ratings were used to calculate **C,** the area under the disease progress curve (AUDPC). Means of bars with the same letters were not different based on Fisher's protected LSD at α=0.05.*

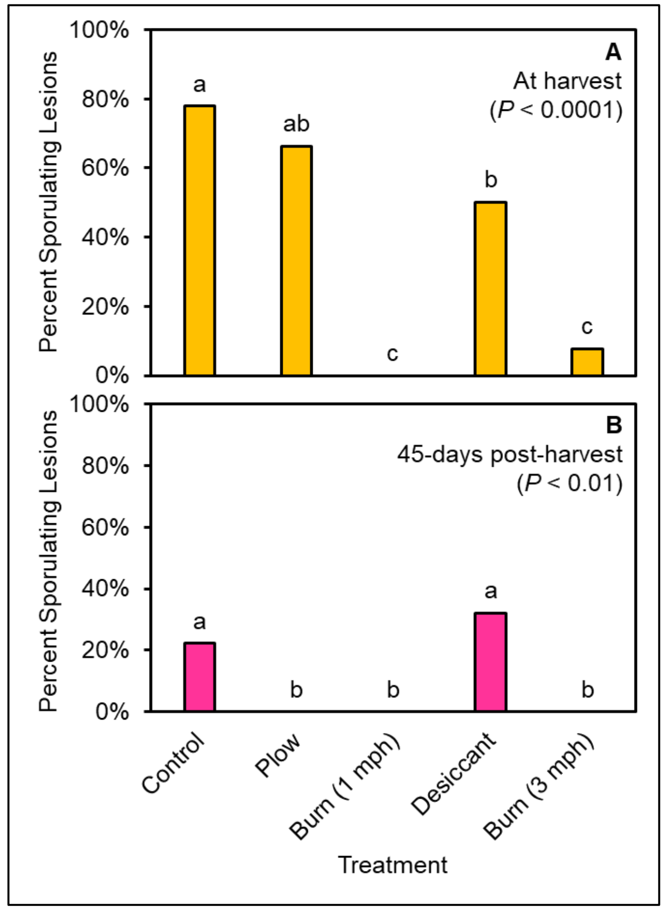

***Figure 2. A,** At-harvest and **B,** 45-days post-harvest lesion sporulation in repeated trial initiated in 2020, following desiccant application 7-days pre-harvest, heat treatment immediately prior to defoliation, and plowing immediately following harvest. Means of bars with the same letters were not different based on Fisher's protected LSD at α=0.05.*