The Potential for secondary Cercosopora leaf spot suppression with foliar fungicide applications made for Rhizoctonia solani management in Sugar Beets

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ABSTRACT

In Wyoming's southeastern irrigated sugar beet production region, field experiments were conducted in the years 2011- 2013 to determine if fungicide applications made for Rhizoctonia root and crown rot disease (RRCR) management would have cross over effect on the later occurring Cercospora leaf spot (CLS) disease development in sugar beet and conversely if later CLS fungicide treatments on their own would have an effect on RRCR development. Secondary goals were to determine if fungicide active ingredient or method of RRCR fungicide application had a differential impact on RRCR and subsequent CLS disease. In 2011, most RRCR-targeted treatments significantly reduced overall CLS disease as measured by AUDPC on average 54% compared to the non-treated check. However, in 2012 and 2013 there was no effect of the early RRCR-targeted treatments on AUDPC for CLS disease. CLS-targeted treatments reduced CLS disease up to 95% but had no effect on RRCR disease. For RRCR, banded fungicide treatments were more effective than broadcast treatments, with no differences in activity between prothioconazole and azoxystrobin. These results provide evidence that fungicide applied for RRCR control can provide some benefit in managing CLS; however, benefits were minor and growers would still be required to use timely fungicide applications to realize economic yields.

INTRODUCTION

Sugar beet producers in the U.S. including the Central High Plains sugar beet production area (Montana, Wyoming, Nebraska, and Colorado), are often plagued with the soil-borne fungal disease Rhizoctonia root and crown rot (RRCR) and the foliar fungal disease Cercospora leaf spot (CLS). These diseases are managed with cultivar selection, crop rotation and reliance on fungicide application targeting each disease individually (Harveson et al., 2009). In addition to the root and crown rot phase, Rhizoctonia can also cause a post-emergence damping-off phase under the proper conditions.

RRCR is caused by Rhizoctonia solani Kühn [Teleomorph: Thanatephorus cucumeris (A.B. Frank) Donk] and is comprised of a number of anastomosis groups with AG-2-2 (which has two subgroups IIIB and IV) capable of causing both disease phases and are the most predominant anastomosis groups in the US beet production areas (Crane et al., 2013; Strausbaugh et al., 2011). For RRCR, the fungus typically initiates infection at the crown spreading over the root surface or can sometimes cause symptoms on the lower part of the root (Harveson et al., 2009). Infection and symptom development can occur from seedling emergence to crown and root infections throughout the growing season (Kirk et al., 2008). Economic losses can be as high as 24% in the U.S. depending on cultivar resistance, environmental conditions and inoculum load (Harveson et al., 2009). RRCR has been identified as the number one disease problem facing growers in the Central High Plains (Larson, 2015).

Management of R. solani includes partially resistant cultivars, fungicide application and agronomic practices (Buhre and Kluth, 2009; Kiewnick et al., 2001; Panella et al., 1994). Since the initial labelling of Quadris® (azoxystrobin) in 1999, growers have multiple fungicide options to manage this disease. These include several strobilurin (QoI), SDHI and DMI fungicides, that can be used in-furrow, or as a foliar-band or broadcast applications to manage this disease (Arabiat and Kahn, 2016; Friskop et al., 2018). Additionally, various seed treatments are available that can provide good early season control (Jacobsen et al., 2012). Because infection can occur over the growing season, research on determining optimal timing for RRCR management has been conducted (Boulton et al., 2010; Jacobsen et al., 2004; Kirk et al., 2008; Stump et al., 2004; Windels and Brantner, 2005). It is generally accepted that for azoxystrobin, application needs to be made before infection occurs (Windels and Brantner, 2005). Foliar applications are targeted to when soil temperatures reach 18 C°, which roughly equates to 4 to 6-leaf beet stage. However, longer season management

may be best achieved with seed or in-furrow treatments followed by foliar-band applications, on up to 12-leaf stages (Jacobsen et al., 2012; Windels and Brantner, 2005; Stump, 2015).

Foliar-band fungicide applications are shown to be more efficacious in managing RRCR than the same fungicide rate applied broadcast (Stump and Franc, 2013). In fact, for most fungicides marketed for RRCR management in sugar beets, product labels indicate the fungicide should be applied as a crop row banded spray. Compared to a broadcast treatment, band applications result in more fungicide active ingredient concentrated near the plant crown and soil near the root zone where infection occur. However, under certain conditions, like low disease pressure, differences between banded and broadcast applications were not always significant (Brantner and Windels, 2012; Franc and Stump, 2010). At the time of this study, some growers in the High plains production region were still applying post-emergent fungicides broadcast since they lacked a band sprayer. Because of this, a comparison of these two application methods were included in the study.

Cercospora leaf spot, caused by the foliar fungal pathogen Cercospora beticola (Sacc.) is considered the most important foliar disease of sugar beet world-wide (Duffus and Ruppel, 1993; Holtschulte, 2000; Weiland and Koch, 2004; Jacobsen and Franc, 2009). CLS disease can result in reduced root yield, lower beet quality, poor beet storage, and lower sucrose content and production losses as high as 40% have been estimated in sugar beet (Smith and Ruppel, 1973; Shane and Teng, 1992). Disease development by C. beticola is favored by daytime temperatures of 25 to 35°C, night temperatures above 16°C, with periods of high humidity or free moisture on leaves for extended periods (Pool and McKay, 1916; Jacobsen and Franc, 2009). In the arid Central High Plains, favorable conditions for CLS infection typically begin at row closure (mid- to late-July) and epidemics can develop until harvest under favorable environmental conditions (Wilson ed, 2001).

Management of CLS includes reduction of inoculum by tillage and crop rotation, cultivar selection, and foliar fungicide applications (Jacobsen and Franc, 2009; Wilson ed, 2001; Miller et al., 1994). Most commercial sugar beet varieties have only moderate resistance at best, therefore growers typically rely on foliar fungicide applications to obtain adequate protection from CLS (Miller et al., 1994). Commonly used fungicides in the High Plains and Minnesota/North Dakota production regions include triphenyltin hydroxide, mancozeb, tetraconazole, prothioconazole, difenoconazole, thiophanate-methyl, pyraclostrobin and commercial mixes of fluxapyroxad + pyraclostrobin

and difenoconazole + propiconazole (Hakk et al., 2016; Secor et al., 2016; Stump, unpublished data). Initiation of fungicide application is typically based on field scouting (starting at row closure) for first lesion development. Subsequent applications are made when symptoms are present and environmental conditions are favorable. Due to fungicide resistance to the majority of these CLS fungicides, tank-mixes with a protectant and fungicide rotations between fungicide class is recommended (Kahn and Hakk, 2016; Secor and Rivera, 2010). In Minnesota and North Dakota, 2 to 7 fungicide applications are made depending on disease pressure (Hakk et al., 2015; Hakk et al., 2016). Since 1998, the University of Wyoming Extension Plant Pathology lab has conducted an annual Cercospora beticola survey for fungicide resistance in the High Plains production region, which includes northeastern Colorado, eastern Montana, western Nebraska and Wyoming (Briere et al., 2001). The past 5 years, growers who submitted samples were queried what and how many fungicide applications were made for CLS and 0-2 applications are typically made for this region (Stump, unpublished survey data).

Coincidentally, many of these fungicides used for RRCR management are also labelled for CLS management in sugar beet. Growers have asked if fungicides applied for RRCR management early in the season would have any carry over effect on CLS disease development.

The objective of this study was to determine if early fungicide applications targeting RRCR would have an effect on the later occurring CLS disease development and conversely if later CLS fungicide treatments on their own would have an effect on RRCR development. Secondary goals were to determine if fungicide active ingredient and method of RRCR fungicide application, foliar broadcast or foliar banded, had an impact on RRCR and subsequent CLS disease. Fungicides selected for the RRCR-targeted treatments, and labeled for both RRCR and CLS, were azoxystrobin (Quadris®, Syngenta Crop Protection LLC, Greensboro, NC) and prothioconazole (Proline®, Bayer CropScience LP, Research Triangle Park, NC). Azoxystrobin was selected because it is considered the industry standard for RRCR management (Hakk et al., 2016). However, though labelled for CLS, azoxystrobin is less effective for CLS management and has resistance problems, and consequently current use is limited (Hakk et al., 2016; Khan and Smith, 2004; Kirk et al., 2012; Stump, unpublished survey data). Prothioconazole has proven to be effective in suppressing both RRCR and CLS (Khan and Hakk, 2016). Fungicides selected for the CLS-targeted treatments included commonly used fungicides, which included prothioconazole.

MATERIALS AND METHODS

The field experiment was conducted at the University of Wyoming Sustainable Agriculture Research and Extension Center (SAREC) located near Lingle, WY in 2011, 2012 and 2013. The elevation of the site is 4165 MSL, and the soil type a Mitchell clay loam at pH = 7.9. Overhead irrigation was applied once a week as needed during the growing season for sugar beet production. Fertilizer was applied as per soil recommendations for sugar beet production and weeds controlled when necessary with applications of glyphosate. For all three years, the field plot was planted with Beta 66RR60, a Roundup Ready cultivar. This cultivar was characterized as having tolerance to Cercospora leaf spot and susceptible to Rhizoctonia solani (BETASEED®, Bloomington, MN). Ideally, a variety with susceptibility to both diseases would have been selected but that was not an option for the Western Sugar Cooperative Southern production region approved varieties. In past experiments, inconsistent Rhizoctonia disease development with tolerant varieties necessitated selection of a variety with susceptibility to Rhizoctonia. The statistical design was a randomized complete block design with four replications; plots were four rows (76.2-cm row centers) by 6.1 m with a 1.5 m in-row buffer. Plants were inoculated with R. solani AG-2-2 and Cercospora beticola to increase disease pressure and reduce in-field variability. For each plot, one of the two middle rows was selected at random, and received Rhizoctonia inoculations. The remaining middle row was utilized for CLS inoculations and evaluations. The RRCR or CLS-targeted fungicide treatments were made to both rows. All data were collected and analyzed from these rows separately. Table 1 summarizes treatment information, Table 2 dates for planting, inoculations, treatment application, and environmental data at fungicide applications. Table 3 summarizes disease evaluation dates and harvest dates for the three years.

INOCULATIONS

Rhizoctonia: Inoculum was prepared using E.G. Ruppel's field inoculum procedure for Rhizoctonia, grain and water amounts were proportionally adjusted to accommodate the size of our stainless-steel trays (Ruppel, 1997). Naturally hull-less barley (3142 cc) was placed in stainless-steel pans (30 cm x 10 cm x 50 cm) with 1886 mL of distilled water and mixed to wet the grain. After absorbing water for 12-14 hours, the pans of grain were autoclaved for 100 min then allowed to cool under a laminar flow hood. Two Rhizoctonia solani AG-2-2 IIIB

isolates (R1 and R9) were provided by Linda Hanson at the USDA-ARS Sugarbeet Research laboratory in Fort Collins, CO. Isolates were recovered from a diseased sugar beet from Colorado and an additional isolate from a diseased sugar beet from Wyoming. Isolates were maintained in an incubator set at 26 C° on full strength Difco® potato dextrose agar prior to inoculation. Each pan was inoculated with one isolate. The hyphae/agar from the plate was aseptically cut into quarters which were individually placed equidistantly in the pan, 5-7 cm below the surface of the grain. Sterile stainless-steel lids were added to the pans and taped into place. Pans were incubated at 25 C° for 3 weeks after which large clumps of infested grain were broken up by hand to facilitate drying and handling. The infested barley grain was spread out over a paper-covered table and air-dried at room temperature for 1 week and then coarsely ground in a Wiley Mill. Subsamples of infested grain from each isolate source were frozen and used to produce fresh inoculum prior to each field season. Inoculum for each of the two isolates were mixed in a 50:50 ratio and stored at 7.2 C° until application.

Immediately following the first fungicide applications, inoculum (3.7 mL = 0.55 g) was applied to the crown of each plant in the designated Rhizoctonia inoculation row for in each plot. Plants were in the 10 to 12-leaf stage each year when inoculated. Shortly after inoculation, plots were cultivated to move soil onto the crown then irrigated (2.54 cm) several hours later to further enhance conditions that favored infection. The author acknowledges that this inoculation methodology can result in consistent but extreme disease pressure, which provides a rigorous test for fungicide efficacy but may not be representative of what a grower would encounter in a typical production year. Stand counts (6.1-row m) were conducted for the Rhizoctonia inoculated rows on dates shown in Table 1.

Cercospora: Cercospora beticola recovered from sugar beets grown at SAREC the previous year were isolated as described in Briere et al. (2001). Resultant colonies were subcultured by hyphal-tip transfers onto amended sugar beet leaf extract agar (SBLA) plates (Ruppel and Hill, 2000). SBLA was prepared by adding 250 g of washed sugar beet leaves (collected the previous year and frozen) to 1 L of distilled water. This mixture was boiled for 40 minutes in a microwave oven, filtered through sterile gauze, and the filtrate volume was adjusted to 1 L with distilled water. Filtrate was amended with 4 g L-1 of glucose (to promote hyphal growth) and 15 g L-1 of water agar and autoclaved for 20 minutes. After cooling, the medium was dispensed into 100 x 15 mm petri plates with an automatic dispensing unit. These cultures were incubated at 22 C° with a 12-hr photoperiod, which resulted in extensive sporulation after one week. After which, the plates were

allowed to dry down for an additional week under the same conditions to facilitate spore removal. This step, kept the agar from tearing as much during plate washing for spore removal. Plates were amended with enough sterile deionized water to cover the bottom of the plate, then gently rubbed with a bent glass rod to dislodge spores. This spore suspension and associated hyphae was filtered through sterile cheesecloth and the spore concentration adjusted with the aid of a hemacytometer to 4 x 103 spores per mL. Foliar inoculations with C. beticola spores and associated hyphae were made to the row not inoculated with R. solani of the two middle rows, and the 1.5-m in-row buffer of each plot. Applications were made when endemic CLS lesions were first observed in the field (Table 2). Inoculum was applied in a total volume of 3.75 L/304.8 m of row via a single-nozzle (8002 flat fan) with the aid of a portable (CO2) sprayer. Efficacy of this method of CLS inoculation was not determined but similar methodologies yielded infection in the field (Adams, et al., 1995).

FUNGICIDE TREATMENTS

Fungicides used in the study timed for RRCR management include Quadris® 2.08 SC (azoxystrobin, 22.9% a.i., Syngenta; Greensboro, NC, USA) and (Proline® 480 SC (prothioconazole 41%, Bayer CropScience LP, Research Triangle Park, NC, USA). In addition to prothioconazole, fungicides timed for CLS included Gem® 500 SC (trifloxystrobin 42.6% a.i., Bayer CropScience LP, Research Triangle Park, NC, USA), Eminent® 125 SL (tetraconazole 11.6% a.i., Sipcam Agro USA, Durham, NC, USA), and Super Tin® 80 WP (triphenytin hydroxide 80% a.i., DuPont, Wilmington, DE, USA). All treatments of prothioconazole also contained the adjuvant Induce® (Alkyl Aryl Polyoxylkane Ethers and Free Fatty Acid, Helena Chemical Co. Memphis, TN). Fungicide treatment descriptions are shown in Table 1 and application dates shown in Table 2. The first application listed for each year is the RRCR banded/broadcast application. The remaining application dates are for CLS management. The RRCR timing application was made one of two ways, as a banded application and as a broadcast application. Both application methods used the same rate per hectare with this amount being concentrated in a band over the row for the banded treatments. Banded fungicide was applied in a 17.8-cm band to plant crowns (10-12-leaf stage) of the two middle rows on dates shown in the Table 1 immediately prior to inoculation. Applications were made via a portable (CO2) sprayer in a total volume of 3.75 L/304.8 m at 0.31 MPa boom pressure. The boom was equipped with a single #8002 flat fan nozzle. Broadcast fungicide, whether for RRCR or CLS, was applied with the aid of a portable (CO2) sprayer in a total volume of 106.3L/ha at 0.207 MPa boom pressure (four #8004 flat fan nozzles spaced at 50.8 cm).

Disease Evaluations RRCR severity in the plots were visually rated for the percentage of total canopy necrosis present. Canopy necrosis was estimated using the Horsfall-Barratt scale (0-11) and ratings were converted to percentage data using the appropriate conversion table for presentation in the Tables (Horsfall and Barratt, 1945). Dates for RRCR disease ratings are shown in Table 3. Foliar necrosis ratings were an indirect estimate of RRCR severity based on foliar collapse.

Cercospora leaf spot severity was determined in each plot (dates in Table 3) on the row not inoculated with Rhizoctonia per plot (but inoculated with C. beticola). The number of lesions on five randomly selected leaves per plot was determined and an average was calculated for each plot. Although more labor intensive than using the KWS visual rating scale (Shane and Teng, 1992), this disease assessment technique can provide fungicide mean separations under low disease pressure years for fungicide efficacy trials (Franc and Stump, 2008). Disease severity data for both RRCR and CLS disease were used to calculate an area under the disease progress curve (AUDPC) rating for each treatment program. The AUDPC is a measure of season-long disease severity for each treatment.

At harvest, beets in each plot were mechanically defoliated with a 4-row defoliator. Beets in the middle two row (the Rhizoctonia inoculated and non-inoculated rows) were then mechanically lifted with a tractor pulled one-row slip, and the beets from the middle 3 m of each row were collected and the total beet root yield was determined with a hanging scale for each row separately. A subsample of approximately 7-10 beets from each plot was submitted to Western Sugar's tare lab for beet quality and percentage of total sucrose determination.

All data were analyzed by analysis of variance (ANOVA, PROC GLM) with four replications using SAS software. Data was combined over years when statistically allowed. Mean separations were done using Fisher's protected LSD ($P \leq 0.05$). For Rhizoctonia evaluations, data were from the Rhizoctonia inoculated row per plot. For CLS treatment evaluations, data were from the row not inoculated with Rhizoctonia per plot.

RESULTS

For all three years, for the Rhizoctonia inoculated row, RRCR development resulted in near total stand loss in the non-treated inoculated check and CLS-targeted treatments by late August (2011-12) or mid-July (2013). However, CLS pressure, despite inoculations,

varied over the years. Based on when disease initiated, final CLS counts and AUDPC values for CLS fungicide trials conducted at this location (2005-2013), CLS disease was considered severe, low and severe in 2011, 2012, and 2013 respectively. For comparison to other beet production areas, the commonly used Kleinwanzleber Saatzucht (KWS) rating was made on the final CLS rating date in 2013, and the non-treated check was 7.3 (Anonymous, 1970). Because CLS and RRCR disease pressure fluctuated by year, there were significant year and year by treatment effects ($P \leq 0.05$) for most parameters measured and years were analyzed separately for all parameters. Study treatment structure and main effects of analysis of variance measured as P values for RRCR and CLS disease severity and extractable sucrose yield for 2011-2013 are shown in Tables 4 and 5.

Sugar beet stands were not significantly different in a given year, meaning stands were relatively consistent across the study area (data not shown). Sugar beet stands overall were low in 2012 compared to 2011 and 2013.

Effects of treatments on overall RRCR disease severity, as measured by the AUDPC for canopy decline, are shown in Table 6 for 2011, 2012 and 2013. Not all evaluation dates are presented in Table 6 for simplicity; the first evaluation and last evaluation are presented along with the computed AUDPC value for each year. Fungicide treatments that targeted RRCR disease reduced the AUDPC of overall canopy decline due to RRCR when compared to the non-treated inoculated check for all three years ($P \leq 0.0001$). In 2011, all RRCR-targeted treatments reduced the AUDPC of canopy necrosis 96-99%, compared to the non-treated inoculated check. However, by the end of the season in 2012 and 2013, considerable disease had developed in the plots throughout the season and levels of disease suppression in the RRCR-targeted treatments ranged from 46 to 84%. Banded treatments were significantly better than the broadcast treatments in reducing overall RRCR AUPDC. Band-applications on average, reduced overall AUDPC 75 and 79%, compared to the 47 and 49% reductions with broadcast treatments in years 2012 and 2013 respectively ($P \leq 0.0001$). However, within a Rhizoctonia–targeted application method there were no differences between prothioconazole and azoxystrobin except on the rating date of 27 August, 2013.

Fungicide treatment programs targeting CLS management (treatments 6-8) when applied to the Rhizoctonia inoculated row, had no effect on RRCR severity, and the tetraconazole program (treatment 8, 2011) even had a greater RRCR AUDPC severity than the non-treated inoculated check ($P \leq 0.0001$). CLS fungicide applications were

initiated and CLS inoculations were made based on row closure timing and first endemic lesion detections. For each year at that time, RRCR infection was well established and the majority of the plants in these CLS-targeted treatments plots were already dead due to RRCR.

Treatment effects on CLS development for 2011-13 are shown in Tables 7-9. In 2011, the three CLS-targeted fungicide programs (treatments 6-8, Table 7) effectively suppressed CLS development compared to the non-treated inoculated check on average 94% and the three treatments were equally effective ($P \leq 0.0001$). Surprisingly, under severe CLS pressure, some RRCR-targeted treatments (treatments 2-5) did significantly reduce CLS disease compared to the non-treated inoculated check on some evaluation dates ($P \leq 0.0001$). Additionally, these treatments reduced the CLS AUDPC compared to the non-treated inoculated check on average 47 % ($P \leq 0.0001$).

In 2012, Cercospora leaf spot development was light, variable, and developed slowly (Table 8). Because of this, there was no significant treatment differentiation for both the individual CLS rating dates and for AUDPC.

Cercospora leaf spot development was severe in 2013 and all CLS-targeted treatments significantly reduced CLS disease similarly on average 82.7% compared to the non-treated inoculated check and the RRCR-targeted treatments (Table 9, $P \leq 0.0001$). For the most part, the RRCR-targeted treatments had little effect on CLS severity with AUDPC values for these treatments not being different from the non-treated inoculated check. However, on 22 August the prothioconazole banded and azoxystrobin broadcast treatment had significantly fewer lesions than the non-treated inoculated check ($P \leq 0.0001$).

Treatment effects on total extractable sucrose yield for the three years are shown in Table 10. Data was partitioned between the Rhizoctonia inoculated rows and rows not inoculated with Rhizoctonia, effectively providing yield effects from Rhizoctonia and CLS disease respectively. In 2011, with Rhizoctonia inoculation, all RRCR-targeted fungicides provided plants early season protection from RRCR resulting greater sucrose yields compared to the non-treated inoculated check, which had no effective yield ($P \leq 0.0001$). The banded azoxystrobin treatment (treatment 3) had on average, 22.6% greater sucrose yields than the two broadcast treatments ($P \leq 0.0001$). The prothioconazole band treatment had an intermediate yield response. The CLS-targeted fungicide programs also effectively had no yields when inoculated with Rhizoctonia and were similar to the non-treated inoculated check.

Though not statistically comparable, sucrose yields for these RRCR-targeted treatments were similar to or slightly less than the yields of these analogous treatments in the absence of RRCR disease (but with limited CLS suppression). Yields in the absence of RRCR disease (data columns for rows inoculated with CLS only) show that CLS-targeted fungicide programs had significantly greater sucrose yields (59% increase on average) than the non-treated inoculated check (P≤0.0011). Although data trends show a slight increase in yields for the RRCR-targeted treatments compared to the non-treated inoculated check, these results were not significant.

In 2012, RRCR development was rapid resulting in poor root yields for the RRCR inoculated row. However, banded applications of prothioconazole and azoxystrobin had greater yields than the non-treated inoculated check, RRCR-targeted broadcast treatments and the CLS-targeted treatments (P≤ 0.0001). For the CLS-targeted treatments in the presence of RRCR there was no harvestable beets at seasons end. Under the conditions of light CLS pressure in 2012, yields in the CLS inoculated rows were statistically similar for all treatments.

In 2013, again under severe RRCR disease pressure, sucrose yields were poor for the RRCR inoculated row. Banded applications of prothioconazole and azoxystrobin had greater yields than the non-treated inoculated check, RRCR-targeted broadcast treatments, and the CLS-targeted treatments (P≤ 0.0001). Sucrose yields for the broadcast prothioconazole and azoxystrobin and CLS-targeted treatments were not significantly better than the non-treated inoculated check. Sucrose yields in the presence of CLS disease show that most CLS-targeted fungicide programs had greater yields than the RRCR-targeted treatments (P≤0.0004). Yields for the RRCR-targeted treatments were not different from the non-treated inoculated check. However, the banded and broadcast prothioconazole treatments (treatments 2 and 4) were statistically similar to the CLS-targeted prothioconazole/triphenyltin hydroxide sequential treatment (Treatment 7). There were no differences between extractable sucrose yields between the CLS-targeted treatments. Though not significantly different, extractable sucrose yields trended higher with RRCR-targeted treatments than the non-treated check lending some support that early RRCR treatments did have some effect on later season CLS disease impacts.

DISCUSSION

The producer driven question regarding if fungicide treatments applied for Rhizoctonia management would have any effect on Cercospora leaf spot management, seems logical since many of the fungicide products utilized have activity against both fungi. Yet, the time disparity between each disease phenology suggest it would be too great for fungicide carryover to have any effect. Most information puts the Quinone outside inhibitors fungicides like azoxystrobin having an effective life in the plant at 7 to 21 days and most DMI fungicides, which include prothioconazole, have a residual period of 14 days (Mueller and Bradley, 2008). The results of this study indicate, in at least one year of the three, under conditions of sprinkler-irrigated Wyoming sugar beet production, there can be some fungicide carryover effect from a 10-12 leaf sugar beet foliar application (azoxystrobin or prothioconazole) resulting in measurable reductions in subsequent CLS disease development. In the 2011 experiment, which was characterized by severe CLS pressure, it was found that early fungicide treatments targeting RRCR did reduce subsequent CLS disease development. However, although overall disease was reduced on average 54%, final yields were not different from the non-treated CLS-inoculated check. In 2012, there were no such effects, most likely due to very low CLS pressure. In 2013, characterized by severe CLS pressure, there was some weak evidence to support that these early Rhizoctonia-targeted fungicides have some additional benefit on CLS management based on significant differences found on one rating date and data trends. These fungicide applications were made prior to row closure and before recommended CLS timings for the High Plains production region (Kahn and Hakk, 2016; Wilson, 2001).

It would be speculation at this point based on the parameters measured in this study, on mechanisms of how these early applications of azoxystrobin or prothioconazole would impact overall CLS development. Both compounds inhibit spore germination and prothioconazole has some limited activity against recent infections. In addition to differences in disease pressure, time between RRCR and CLS treatments was 27 days in 2011 and much longer in 2012 and 2013 (41 and 44 days respectively) which may account for differences seen in CLS management. Since the incubation period of C. beticola development is 7-21 days, depending on environmental conditions, it is plausible that these early fungicide applications could be targeting primary inoculum, thereby delaying disease development.

For the three years the study was conducted fungicide treatment programs targeting CLS management had no significant effect on RRCR suppression because fungicide was applied too late. Infection was already present and many beets were already dead when fungicide was applied. Because of the severe RRCR disease pressure, this study could not address whether these CLS-targeted fungicide treatments would have any effect on RRCR under a more moderate disease pressure situation.

Banded fungicide treatments, with the exception in the 2011 study, were found to be superior to broadcast treatments for RRCR suppression and sucrose yield ($P \leq 0.0001$). Averaging over the 2012 and 2013 studies, banded fungicides reduced RRCR overall disease 77% vs. 48% when compared with the same fungicides applied broadcast. These findings agree with previous direct comparisons of banded vs. broadcast treatments conducted in Wyoming (Stump, 2013). Banding of fungicide effectively concentrates a broadcast amount in a 12-18 cm band delivering more active ingredient for plant uptake.

Fungicide efficacy for suppressing RRCR was similar between prothioconazole and azoystrobin. These findings also agree with numerous field trials conducted by the author (unpublished data). In the High Plains production region, azoystrobin has been the industry standard since it was introduced, but it is useful for growers to have additional viable options for RRCR management, especially in resistance management. Though no R. solani involved with RRCR has been found with resistance to azoxystrobin, Arabiat and Kahn (2016) demonstrated that exposure of R. solani AG-2-2 populations to azoxystrobin in vivo and vitro studies resulted in isolate shifts to reduced sensitivity.

Despite some potential beneficial effects on supressing CLS disease in Wyoming, a grower would not be advised to depend on these treatments solely for CLS management, as impacts, though significant at times, were relatively minor. Future research should address whether the combination of RRCR-targeted fungicide applications followed with CLS-targeted applications would have less overall CLS disease then just CLS-targeting treatments. Additionally, it would be of interest to investigate how RRCR management programs of seed treatment or in-furrow fungicide application, which allow for later beet growth stage banded applications (10 to 12-leaf), would affect CLS disease.

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Table 1. Descriptions of banded and broadcast fungicide applications for Rhizoctonia root and crown rot and Cercospora leaf spot management in sugar beet, Lingle, WY experiments conducted in 2011-13.

Treatment designation	**Treatment and application rate**	**Application timing**
1. Non-treated inoculated check	1. Inoculated, no fungicide	NA
2. Early season banded Prothioconazole targeting Rhizoctonia	2. Prothioconazole (4.64 g ai/304.8 m) + Induce (0.125% v:v)	10-12 leaf-stage
3. Early season banded Azoxystrobin targeting Rhizoctonia	3. Azoxystrobin (4.44 g ai/304.8 m)	10-12 leaf-stage
4. Early season broadcast Prothioconazole targeting Rhizoctonia	4. Prothioconazole (20.0 g ai/ha) + Induce (0.125% v:v)	10-12 leaf-stage
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	5. Azoxystrobin (18.3 g ai/ha)	10-12 leaf-stage
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14 day intervals targeting CLS	6. Prothioconazole (17.5 g ai/ha) + Induce (0.125% v:v).... 6. Trifloxystrobin (13.2 g ai/ha) 6. Triphenyltin Hydroxide (21 g ai/ha)	At start of CLS initiation 14-days after CLS initiation 28 days after CLS initiation
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14 day intervals targeting CLS	7. Prothioconazole (17.5 g ai/ha) + Induce (0.125% v:v).... 7. Triphenyltin Hydroxide (21 g ai/ha)	At start of CLS initiation 14-days after CLS initiation
8. Tetraconazole broadcast 3 times at 14 day intervals targeting CLS	8. Tetraconazole (11.4 gai/ha)	At start of CLS initiation 14-days after CLS initiation 28 days after CLS initiation

Table 2. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot and Cercospora leaf spot management in sugar beet, Lingle, WY. Environmental conditions and plant growth phase at time of treatment applications.

Year	Planting date	Fertility	Inoculation dates		Treatment application dates	Air Temperature (C°)	Wind speed (kph)
			Rhizoctonia solani	*Cercospora beticola* (endemic disease appearance/inoculation)			
2011	29 April	27.2 Kg N + 22.7 Kg P_2O_5 + 9.1 Kg S on 4 April; 13.6 Kg N + 13.6 Kg P_2O_5 + 4.5 Kg S on 25 April	30 June	27 July/27 July	30 June 27 July 11 August 24 August	23.3 32.2 20.0 31.7	0 0 0 8
2012	4 May	45.4 Kg N + 34 Kg P_2O_5 + 9.1 Kg S on 11 April	21 June	8 August/16 August	21 June 1 August 17 August 29 August	22.2 21.1 22.8 17.2	10 0 11 0
2013	7 May	22.7 Kg N + 20.4 Kg P_2O_5 + 9.1 Kg S on 29 March and 22.7 Kg N + 11.3 Kg P_2O_5 on 30 April	18 June	1 August/15 August	18 June 1 August 12 August 27 August	23.3 32.2 21.1 18.9	8 0 0 0

Table 3. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet, Lingle, WY. RRCR, and CLS disease evaluations and harvest dates in sugar beet 2011-13

Year	**RRCR disease severity evaluations dates**	**CLS disease severity evaluation dates**	**Harvest dates**
2011	15 July 28 July 10 August 24 August	10 August 17 August 24 August 1 September 8 September	4 October
2012	17 July 8 August 28 August 12 September	8 August 17 August 28 August 5 September 12 September	24 September
2013	3 July 17 July 27 August	6 August 15 August 22 August 26 August	1 October

Table 4. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot and Cercospora leaf spot management in sugar beet, Lingle, WY. Main effects of analysis of variance measured as *P* values for RRCR and CLS disease severity for 2011-2013.

Source	df	RRCR AUDPC[x]			CLS AUDPC[y]		
		2011	2012	2013	2011	2012	2013
Model	10	<0.0001	<0.0001	<0.0001	<0.0001	0.1143	<0.0001
Treatment	7	<0.0001	<0.0001	<0.0001	<0.0001	0.3089	<0.0001
Rep	3	0.9365	0.5756	0.3755	0.0292	0.0458	0.1648
Error	21	…	…	…	…	…	…

[x] The AUDPC (area under the disease progress curve) for canopy necrosis data collected for 15 Jul to 24 Aug (2011), 17 Jul to 12 Sep (2012), and 3 Jul to 27 Aug 92013).

[y] The AUDPC (area under the disease progress curve) for CLS lesion data collected for 27 Jul to 8 Sep (2011), 8 Aug to 12 Sep (2012), and 6 to 26 Aug (2013).

Table 5. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot and Cercospora leaf spot management in sugar beet, Lingle, WY. Main effects of analysis of variance measured as *P* values for extractable sucrose yield for 2011-2013.

Source	df	2011		2012		2013	
		Rhizoc row yield	CLS row yield	Rhizoc row yield	CLS row yield	Rhizoc row yield	CLS row yield
Model	10	<0.0001	0.0037	<0.0001	0.0713	<0.0001	0.0009
Treatment	7	<0.0001	0.0011	<0.0001	0.1447	<0.0001	0.0004
Rep	3	0.0097	0.6331	0.2107	0.0593	0.2107	0.2381
Error	21	…	…	…	…	…	…

Table 6. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet, Lingle, WY. Fungicide treatment effects on RRCR in the RRCR inoculated row.

Treatment designation[z]	RRCR severity as a percentage of total canopy necrosis and AUDPC[y]								
	2011			2012			2013		
	15 Jul	24 Aug	AUDPC	17 Jul	12 Sep	AUDPC	3 Jul	27 Aug	AUDPC
1. Non-treated inoculated check	1.6 b[x]	97.0 a	275.0 b	50.0 b	97.0 a	512.9 b	23.5 a	100.0 a	518.1 a
2. Early season banded Prothioconazole targeting Rhizoctonia	0.0 b	3.0 b	8.8 c	0.6 c	17.0 d	110.8 d	0.0 b	23.5 e	82.0 c
3. Early season banded Azoxystrobin targeting Rhizoctonia	0.0 b	1.0 b	3.5 c	0.6 c	28.0 cd	150.0 d	0.0 b	59.5 d	136.8 c
4. Early season broadcast Prothioconazole targeting Rhizoctonia	0.0 b	5.0 b	12.3 c	0.6 c	65.0 b	267.9 c	0.0 b	97.0 b	282.4 b
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	0.0 b	3.0 b	8.8 c	83.0 c	59.5 bc	279.5 c	0.6 b	88.0 c	249.8 b
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14-day intervals targeting CLS	1.6 b	99.0 a	293.6 ab	69.0 ab	100.0 a	613.4 ab	21.0 a	99.5 ab	509.8 a
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14-day intervals targeting CLS	2.0 ab	98.3 a	291.1 ab	88.0 a	100.0 a	643.5 a	17.0 a	99.5 ab	506.5 a
8. Tetraconazole broadcast 3 times at 14-day intervals targeting CLS	6.0 a	99.0 a	332.6 a	59.5 ab	99.0 a	591.6 ab	23.5 a	100.0 a	538.8 a

[z] Treatment description, rates and timing information are described in Table1.

[y] The AUDPC (area under the disease progress curve) for data collected for 15 Jul to 24 Aug (2011), 17 Jul to 12 Sep (2012), and 3 Jul to 27 Aug 92013).

[x] Treatment means followed by different letters in a column differ significantly (Fisher's protected LSD, $P \leq 0.05$).

Table 7. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet, Lingle, WY. Fungicide treatment effects on CLS development, 2011.

Treatment designation[z]	Ave number of lesions per leaf					AUDPC[y]
	10 Aug	17 Aug	24 Aug	1 Sep	8 Sep	
1. Non-treated inoculated check	13.2 a[x]	42.0 a	79.2 a	159.7 a	322.9 a	3234.5 a
2. Early season banded Prothioconazole targeting Rhizoctonia	7.0 ab	16.1 a	18.1 c	83.0 b	192.7 b	1567.8 b
3. Early season banded Azoxystrobin targeting Rhizoctonia	0.5 bc	4.4 bc	14.1 c	87.3 b	210.0 ab	1480.3 b
4. Early season broadcast Prothioconazole targeting Rhizoctonia	1.7 bc	8.4 abc	22.8 bc	106.7 ab	176.5 b	1600.0 a
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	1.9 bc	10.2 ab	43.6 b	75.3 b	151.3 b	1451.9 b
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14-day intervals targeting CLS	0.0 c	0.1 d	2.9 c	13.8 c	12.1 c	159.6 c
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14-day intervals targeting CLS	0.4 bc	0.3 d	8.9 c	12.2 c	12.7 c	197.1 c
8. Tetraconazole broadcast 3 times at 14-day intervals targeting CLS	0.3 bc	3.5 cd	7.4 c	9.8 c	11.4 c	187.3 c

[z] Treatment description, rates and timing information are described in Table1.

[y] The AUDPC (area under the disease progress curve) for data collected for 27 Jul to 8 Sep (27 Jul data not presented in table).

[x] Treatment means followed by different letters in a column differ significantly (Fisher's protected LSD, $P \leq 0.05$).

Table 8. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet, Lingle, WY. Fungicide treatment effects on CLS development, 2012.

Treatment designation[z]	**Ave number of lesions per leaf**					**AUDPC**[y]
	8 Aug	**17 Aug**	**28 Aug**	**5 Sep**	**12 Sep**	
1. Non-treated inoculated check	0.0 a[x]	0.7 a	1.9 a	1.1 a	10.9 a	70.4 a
2. Early season banded Prothioconazole targeting Rhizoctonia	0.0 a	1.1 a	2.3 a	2.6 a	19.1 a	118.7 a
3. Early season banded Azoxystrobin targeting Rhizoctonia	0.0 a	0.2 a	4.2 a	10.5 a	6.2 a	141.7 a
4. Early season broadcast Prothioconazole targeting Rhizoctonia	0.0 a	0.2 a	5.8 a	5.5 a	3.9 a	111.5 a
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	0.1 a	0.7 a	2.6 a	1.4 a	9.1 a	73.4 a
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14-day intervals targeting CLS	0.1 a	0.0 a	0.8 a	5.5 a	3.7 a	61.6 a
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14-day intervals targeting CLS	0.1 a	0.1 a	0.3 a	1.6 a	2.3 a	23.9 a
8. Tetraconazole broadcast 3 times at 14-day intervals targeting CLS	0.1 a	0.2 a	0.6 a	0.2 a	0.5 a	10.8 a

[z] Treatment description, rates and timing information are described in Table1.

[y] The AUDPC (area under the disease progress curve) for data collected for 8 Aug to 12 Sep.

[x] Treatment means followed by different letters in a column differ significantly (Fisher's protected LSD, $P \leq 0.05$).

Table 9. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet, Lingle, WY. Fungicide treatment effects on CLS development, 2013.

Treatment designation[z]	Ave number of lesions per leaf				AUDPC[y]
	6 Aug	15 Aug	22 Aug	26 Aug	
1. Non-treated inoculated check	16.9 bc[x]	149.2 a	219.3 a	292.5 a	3102.3 ab
2. Early season banded Prothioconazole targeting Rhizoctonia	51.8 a	120.1 a	131.1 b	316.1 a	2675.9 b
3. Early season banded Azoxystrobin targeting Rhizoctonia	39.9 ab	182.3 a	231.4 a	376.7 a	3763.1 a
4. Early season broadcast Prothioconazole targeting Rhizoctonia	26.4 a-c	150.3 a	177.4 ab	354.8 a	3072.2 ab
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	20.4 bc	114.5 a	141.9 b	312.0 a	2462.8 b
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14-day intervals targeting CLS	24.2 bc	16.3 b	8.9 c	39.2 b	426.0 c
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14-day intervals targeting CLS	8.4 c	43.5 b	22.3 c	83.8 b	696.2 c
8. Tetraconazole broadcast 3 times at 14-day intervals targeting CLS	10.0 c	43.3 b	4.3 c	26.3 b	492.3 c

[z] Treatment description, rates and timing information are described in Table1.

[y] The AUDPC (area under the disease progress curve) for data collected for 6 to 26 Aug.

[x] Treatment means followed by different letters in a column differ significantly (Fisher's protected LSD, $P \leq 0.05$).

Table 10. Banded and broadcast fungicide applications for Rhizoctonia root and crown rot (RRCR) and Cercospora leaf spot (CLS) management in sugar beet. Fungicide treatment effects on yield. Lingle, WY 2011-13.

Treatment designation[z]	Total extractable sucrose (Kg/ha)					
	2011		2012		2013	
	Data for Rhizoctonia inoculated rows	Data for rows inoculated with CLS	Data for Rhizoctonia inoculated rows	Data for rows inoculated with CLS	Data for Rhizoctonia inoculated rows	Data for rows inoculated with CLS
1. Non-treated inoculated check	0.0 c	5459.0 b	0.0 c	6934.0 a	0.0 b	3605.9 c
2. Early season banded Prothioconazole targeting Rhizoctonia	5626.3 ab	6324.2 b	3658.9 a	5514.0 a	1920.8 a	4776.1 bc
3. Early season banded Azoxystrobin targeting Rhizoctonia	6480.9 a	6319.0 b	4712.7 a	5303.0 a	1467.2 a	4268.2 c
4. Early season broadcast Prothioconazole targeting Rhizoctonia	4732.7 b	6563.2 b	1105.0 b	5128.0 a	186.1 b	4608.2 bc
5. Early season broadcast Azoxystrobin targeting Rhizoctonia	5301.1 b	6284.1 b	627.6 b	4070.0 a	335.0 b	4481.0 c
6. Sequential broadcast fungicide program of Prothioconazole/trifloxystrobin/triphenyltin hydroxide at 14-day intervals targeting CLS	0.0 c	9015.8 a	0.0 c	7189.0 a	0.0 b	6538.3 a
7. Sequential broadcast fungicide program of Prothioconazole/triphenyltin hydroxide at 14-day intervals targeting CLS	322.5 c	8510.8 a	0.0 c	7063.0 a	0.0 b	5899.6 ab
8. Tetraconazole broadcast 3 times at 14-day intervals targeting CLS	0.0 c	8496.4 a	0.0 c	6089.0 a	76.6 b	6789.7 a

[z] Treatment description, rates and timing information are described in Table1.

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