

# EFFECT OF *RHIZOCTONIA SOLANI* INOCULUM DENSITY AND SUGARBEET VARIETY SUSCEPTIBILITY ON DISEASE ONSET AND DEVELOPMENT

Jason R. Brantner, Hal R. Mickelson, and Elizabeth A. Crane

Research Fellow, Scientist, and Junior Scientist, respectively, University of Minnesota, Northwest Research and Outreach Center, Crookston, MN 56716

Rhizoctonia diseases (seedling damping-off and crown and root rot, RCRR), caused by *Rhizoctonia solani* AG 2-2, continue to be among the most common problems on sugarbeet in the Red River Valley and southern Minnesota. Fungicides are available for in-furrow and postemergence applications for control of *Rhizoctonia*, but questions continue to arise about the timing of postemergence applications. Azoxystrobin (Quadris) is effective against RCRR in sugarbeet when applied prior to infection, but is less effective or ineffective after infections have occurred (11). Thus, knowing when infections begin to occur (disease onset) is critical to making timely, effective postemergence fungicide applications.

Rhizoctonia crown and root rot is influenced by soil temperature and moisture. Bolton et al. (1) found that a daily accumulation of 11 growing degree days (GDD) was necessary for infection, and disease developed at a soil moisture holding capacity as low as 25% with enhanced development as soil moisture levels increased. Several studies have evaluated the effect of soil temperature at application of azoxystrobin on control of RCRR (5, 6, 7, and 8). Applications of azoxystrobin at 4-inch soil temperatures ranging from 50 to 73°F resulted in statistically equal disease control and recoverable sucrose per acre, but application at 62 to 67°F tended to give best results in 2003 and 2004 (6, 7). This has led to the adoption of a 60-65°F 4-inch soil temperature threshold for applying postemergence fungicides for control of RCRR. However, this threshold is often reached before sugarbeet seedlings emerge, or shortly after emergence when there is not much foliage present for making a postemergence application. In addition, results have not always been consistent. In 2005, Jacobsen et al. (5) reported significant control with azoxystrobin applications at 4-inch soil temperatures up to 80°F, which was higher than in previous years. In Michigan, soil temperature thresholds did not improve efficacy of azoxystrobin applications, and the authors found planting date, seedling development, or leaf stage more reliable indicators of when to apply fungicides (8).

While soil temperature and moisture are clearly important in infection and development of RCRR on sugarbeet, other factors, such as inoculum density and variety resistance also may play an important role. There are examples of these in other crops. For the soilborne pathogen *Verticillium dahliae*, higher inoculum densities resulted in earlier disease onset in cauliflower compared to lower inoculum densities (12). Similarly for Fusarium wilt of chickpea, increasing inoculum density of *F. oxysporum* caused an exponential reduction in disease incubation period (9). In peanut, planting moderately resistant varieties delayed onset of epidemics of *Cylindrocladium* black rot (4).

## OBJECTIVES

A field trial was established to evaluate the effect of *R. solani* inoculum density and sugarbeet variety susceptibility on onset and development of Rhizoctonia damping-off and crown and root rot.

## MATERIALS AND METHODS

The trial was established at the University of Minnesota, Northwest Research and Outreach Center, Crookston. A factorial set of treatments (*R. solani* inoculum density x variety susceptibility x irrigation) was set up in a split-plot design with four replicates. Inoculum density treatments included 0, 20, 40, and 60 kg ha<sup>-1</sup> *R. solani*-infested whole barley grain broadcast in plots and worked into the top 4 inches of soil with a Melroe multiweeder prior to planting on May 9. Varieties included a partially resistant, moderate, and susceptible. Mean Rhizoctonia root rot ratings from 2011 American Crystal Sugar Company tests were 3.5, 4.7, and 5.3 for the resistant, moderate, and susceptible varieties, respectively. Seed was sown at a 4.5-inch spacing into 6-row plots that were 25 ft long with 22-inch row spacing. Counter 20G (9 lb A<sup>-1</sup>) was applied at planting for control of root maggot and 22 oz A<sup>-1</sup> glyphosate (4.5 lb product ae/gallon) was applied June 13 and July 8 for control of weeds. Plots were split and rows 2 and 3 were

irrigated with trickle-tape for 4.5, 5, 6, 4, 4, 4.5, and 4 hr on July 12, 16, 19, 26, August 1, 13, and 22, respectively. *Cercospora* leafspot was controlled by application of 9 oz Headline in 20 gallons of water A<sup>-1</sup> with a tractor-mounted sprayer with TeeJet 8002 flat fan nozzles at 100 psi on August 23.

Soil samples were taken in both irrigated and non-irrigated rows of two replicates (4 samples/replicate) from the top 4 inches weekly from May 31 to September 5 to calculate percent soil moisture, and 4-inch soil temperature and moisture (kPa) were recorded at 1-hour intervals throughout the growing season. The center four rows of each plot were counted once or twice weekly beginning 14 days after planting through September 5.

The center two rows of plots were harvested September 23. Data were collected for number of harvested roots, yield, and quality. Row 3 data was used to represent irrigated rows and row 4 data was used for non-irrigated rows. Ten roots from each of rows 3 and 4 also were arbitrarily selected and rated for severity of RCRR using a 0 to 7 scale (0 = healthy root, 7 = root completely rotted and foliage dead).

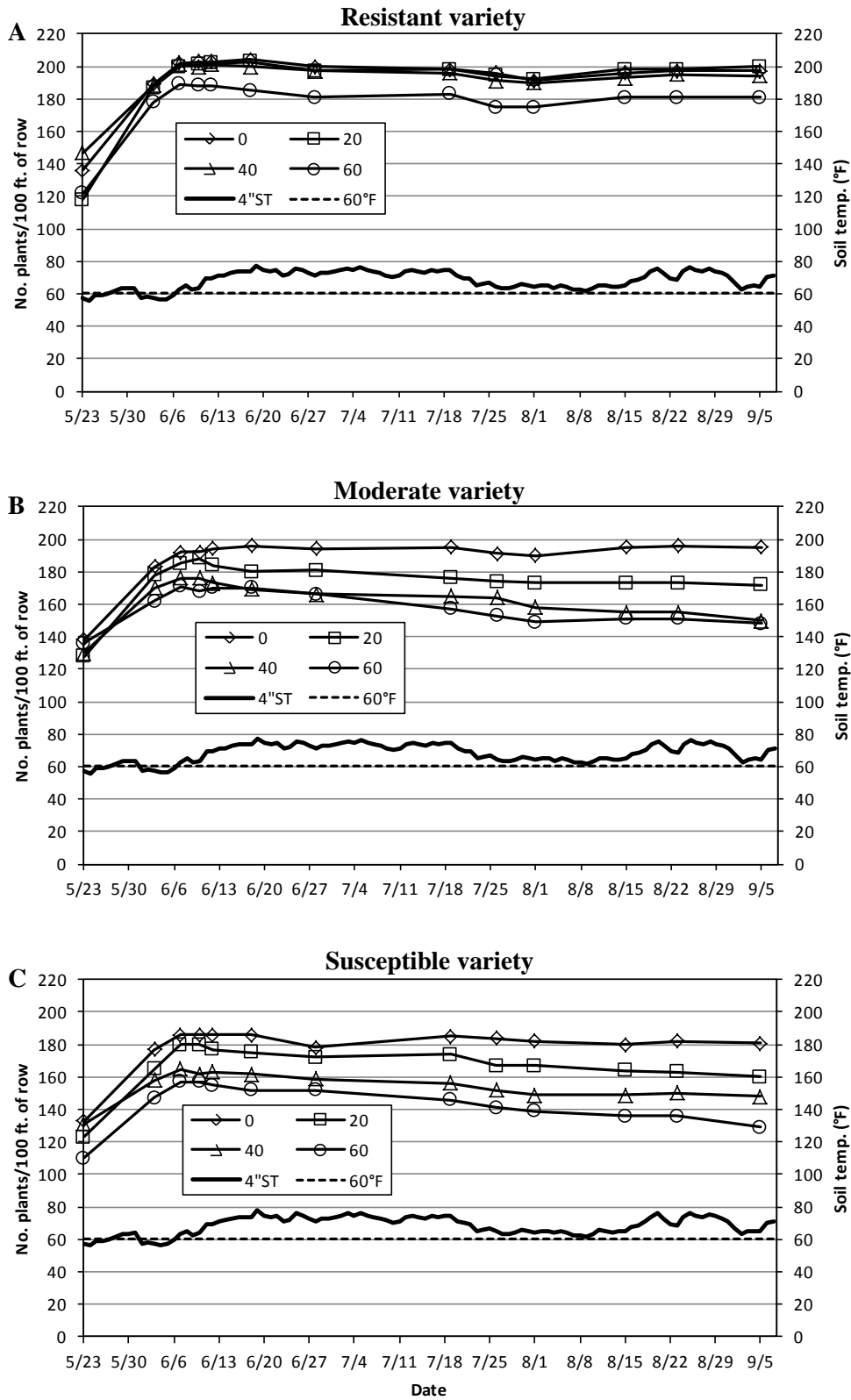
Data were subjected to analysis of variance and orthogonal polynomial contrasts for comparison of main effects of inoculum density, variety susceptibility, and irrigation, and all possible interactions using SAS Proc GLM (SAS Institute, Cary, NC).

## RESULTS

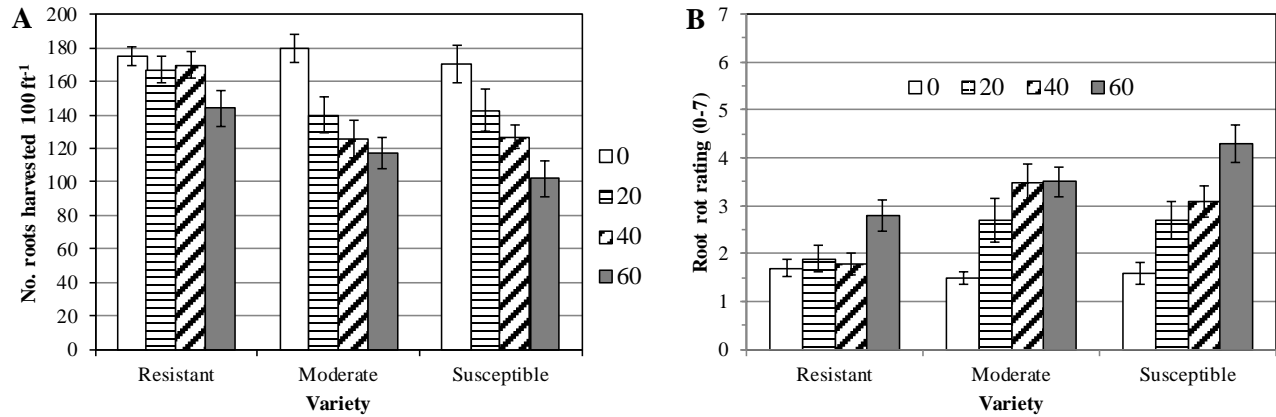
Analysis of stand count data demonstrated significant ( $P = 0.05$ ) linear effects for both inoculum density and variety susceptibility for all stand counts beginning June 3 (3 ½ weeks after planting) and significant ( $P = 0.05$ ) inoculum density by variety interactions at most stand count dates. Inoculum density by variety interactions for stand are summarized in Fig. 1. For all varieties, emergence reached its highest by June 7. Also, by June 7, the daily mean 4-inch soil temperature surpassed 60°F and stayed above that threshold through the rest of the stand count dates. For the resistant variety (Fig. 1A), emergence and stands were similar for inoculum densities of 0, 20, and 40 kg ha<sup>-1</sup> and lower for the 60 kg ha<sup>-1</sup> inoculum density. For the moderate and susceptible varieties (Fig. 1B and C), plots inoculated with 20, 40, and 60 kg ha<sup>-1</sup> demonstrated stepwise reduction in emergence by June 7 and stand throughout the season. Stand for the resistant variety remained steady, while both susceptible and moderate varieties slowly lost stand throughout the season in inoculated plots (Fig. 1). The interaction of inoculum density and variety on stand data is also illustrated for the number of harvested roots per 100 ft of row in Fig. 2A. There was a significant linear effect of inoculum density on all three varieties, but the effect was greater for the moderate and susceptible variety than for the resistant variety, which had a flatter slope (Fig. 2A). Irrigation did not significantly affect stand counts (data not shown).

There were significant inoculum density by variety interactions for *Rhizoctonia* crown and root rot rating at harvest (illustrated in Fig. 2B), yield, and recoverable sucrose A<sup>-1</sup> (Table 1). Root rot rating of the resistant variety did not increase until the inoculum density was 60 kg ha<sup>-1</sup> while root rot ratings for both the moderate and susceptible varieties increased as inoculum density increased from 0 to 60 kg ha<sup>-1</sup> (Fig. 2B). There was a significant linear reduction in both yield and recoverable sucrose A<sup>-1</sup> as inoculum density increased for both the moderate and susceptible varieties, but the effect was not significant ( $P = 0.05$ ) for the resistant variety (Table 1). There was also a significant main effect of inoculum density and significant variety by irrigation interaction on percent sugar (Table 1 and Fig. 3, respectively). Percent sucrose decreased in inoculated compared to non-inoculated plots across all varieties (Table 1). Irrigation increased percent sucrose of the resistant variety, had no effect on the moderate variety, and reduced percent sucrose of the susceptible variety (Fig. 3). Irrigation did not significantly ( $P = 0.05$ ) affect other harvest parameters (data not shown).

Mean 4-inch soil temperatures were above 60°F beginning May 28 (19 days after planting), dipped below 60°F from June 1-6, and then continued to stay above 60°F for the rest of the growing season (Fig. 1). Soil moisture was high early in the season at 30% on May 31, but slowly dropped to 16% by July 10 prompting the irrigation of subplots (which began on July 12). Soil moisture in non-irrigated rows remained low, ranging from 10-17% from July 19 to August 23, up to 22% on August 30, and back down to 15% on September 5. In irrigated rows, soil moisture ranged from 21-25% from July 19 to August 30 and dropped down to 17% on September 5.



**Fig. 1.** Plant stand for sugarbeet **A)** resistant, **B)** moderately susceptible, and **C)** susceptible to *Rhizoctonia* crown and root rot sown in plots infested with *Rhizoctonia solani* at inoculum densities of 0, 20, 40, and 60 kg ha<sup>-1</sup> and mean 4-inch soil temperature (4''ST) in a field trial sown May 9. The dotted line shows 60 °F soil temperature threshold for favorability for *R. solani*-infection. Each symbol for the stand counts represents the mean of 8 plots (four replicate plots across two irrigation treatments). For most stand count dates beginning June 7, there was a significant ( $P = 0.05$ ) inoculum rate by variety interaction.



**Fig. 2.** Interaction of sugarbeet variety (resistant, moderately resistant, and susceptible to *Rhizoctonia solani*) and *R. solani* inoculum density (0, 20, 40, and 60 kg ha<sup>-1</sup>) on **A**) number of harvested roots and **B**) *Rhizoctonia* crown and root rot rating in a field trial sown May 9. Bars in **A** represent the mean of 8 plots (4 replicate plots across irrigated and non-irrigated subplots). Bars in **B** represent the mean of 80 roots (10 roots per subplot x 4 replicate plots across 2 irrigation treatments). For both **A** and **B**, error bars are plus and minus one standard error. Linear effect of inoculum density on the number of harvested roots and root rot rating was significant at  $P = 0.05$  for the resistant variety and at  $P = 0.001$  for the moderate and susceptible varieties.

**Table 1.** Yield, percent sucrose, and recoverable sucrose A<sup>-1</sup> for sugarbeet sown May 9, 2013 with resistant, moderate, and susceptible varieties in a field infested at various inoculum densities of *Rhizoctonia solani*.

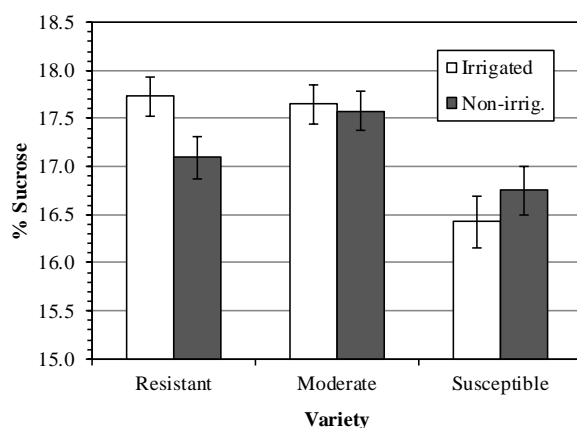
Inoculum rate <sup>y</sup>	Yield (ton A <sup>-1</sup> ) <sup>w</sup>			% Sucrose <sup>x</sup> Across all varieties	Recoverable sucrose (lb A <sup>-1</sup> ) <sup>w</sup>		
	Res.	Mod.	Susc.		Res.	Mod.	Susc.
0 kg ha <sup>-1</sup>	29.1	29.6	33.5	17.6	9621	9959	10787
20 kg ha <sup>-1</sup>	27.0	23.6	28.7	17.0	8831	7810	8663
40 kg ha <sup>-1</sup>	27.8	23.8	30.2	17.1	9006	7745	9344
60 kg ha <sup>-1</sup>	25.5	19.3	22.9	17.0	8294	6394	6820
Linear <sup>z</sup>	NS	***	***	**	NS	***	***
Quadratic <sup>z</sup>	NS	NS	NS	NS	NS	NS	NS
Inoc x Vty <sup>wxz</sup>		*		NS		*	

<sup>w</sup> There were significant interactions between inoculum rate linear response and variety for yield and recoverable sucrose so response to inoculum rate is shown separately for each variety. Values represent the mean of 8 plots (4 replicate plots across 2 irrigation treatments).

<sup>x</sup> There was no significant interaction between inoculum rate linear response and variety for percent sucrose, so response to inoculum rate is shown across varieties. Values represent the mean of 24 plots (4 replicate plots across 3 varieties and 2 irrigation treatments).

<sup>y</sup> *Rhizoctonia solani*-infested whole grain barley inoculum was broadcast in main plots and worked into the top 4 inches of soil with a Melroe multiweeder prior to planting.

<sup>z</sup> Response to inoculum rate and inoculum by variety interactions were tested using orthogonal polynomial contrasts; \* = significant at  $P = 0.05$ , \*\* = significant at  $P = 0.01$ , \*\*\* = significant at  $P = 0.001$ .



**Fig. 3.** Interaction of sugarbeet variety (resistant, moderately susceptible, and susceptible to *Rhizoctonia solani*) and irrigation treatment (irrigated or non-irrigated) on percent sucrose of sugarbeet sown May 9, 2013 in a field trial infested with various inoculum densities of *Rhizoctonia solani*. Bars represent the mean of 16 plots (4 replicate plots across 4 inoculum densities). Error bars are plus and minus one standard error.

## DISCUSSION

Early-season disease pressure was reduced by the lack of concurrent favorable soil moisture and temperature conditions. Soil moisture was high early resulting in excellent emergence. While soil moistures were high and favorable for disease, soil temperatures were low, reducing the development of damping-off. Mean 4-inch soil temperatures hovered right around 60°F, climbing above 60°F around June 7. At this time soil moisture was declining, from 30% on May 31 down to 16% on July 10. Irrigation of subplots began on July 12, so that by July 19 soil moisture was up to 22% in irrigated subplots and only 13% in non-irrigated subplots. By this time, plants were larger and not as susceptible to rapid damping-off; there was no significant effect of irrigation on plant stands. In an inoculated field trial in 2010, inoculum density of 35 kg ha<sup>-1</sup> *R. solani*-infested barley resulted in ~30% stand reduction over 4 weeks compared to non-inoculated plots (3). By comparison, in this trial, stand reduction from the non-inoculated plots for the susceptible variety after 4 weeks was 3, 11, and 16% for inoculum densities of 20, 40, and 60 kg ha<sup>-1</sup>, respectively. Last year in this trial (2), stand reduction from the non-inoculated plots for the susceptible variety after 4 weeks was 0, 13, and 6% for inoculum densities of 20, 40, and 60 kg ha<sup>-1</sup>, respectively.

A major objective of this trial was to determine the onset of disease for the different inoculum densities and varieties. In this trial, there was no particular date that could be determined as the onset of disease. Instead, increased inoculum density resulted immediately in reduced emergence and stand, and this effect of inoculum rate on stand gradually accumulated throughout the growing season, culminating in a highly significant linear effect on harvest parameters (Table 1). These immediate and cumulative effects of inoculum density on emergence, stand, and harvest emphasize the importance of full-season control, including an at-planting treatment such as seed or in-furrow fungicide. These results, however, do not explain observations in some of our past field trials and in growers' fields where disease has not begun until late in the season and at-planting treatments have not provided significant benefit. Perhaps pathogen populations in some growers' fields were lower than the lowest rate in this trial and took time to increase before reaching a level where sugarbeet infection occurred. Additionally, soil type, previous crop residue, or other environmental factors may influence disease onset.

Similarly, the susceptibility of the sugarbeet variety to *Rhizoctonia* affected emergence and stand throughout the growing season. Emergence was complete by 4 weeks after planting, and the resistant variety had highest stand followed by the moderate variety, while the susceptible variety had lowest stand. Even though these stand differences remained throughout the growing season, yield and recoverable sucrose were comparable for the susceptible and resistant varieties until inoculum density reached 60 kg ha<sup>-1</sup>. This is likely due to the very high yield potential of the susceptible variety compared to the other two varieties (see yield for the 0 kg ha<sup>-1</sup> inoculum rate in Table 1). In American Crystal Sugar Company's 2010 and 2011 official variety trials, the two-year mean for yield of the resistant, moderate, and susceptible varieties was 26.0, 26.0, and 29.0 ton A<sup>-1</sup>, respectively (10).

In this trial, sugarbeet varieties differing in susceptibility to *R. solani* responded differentially to inoculum density, based on the significant interaction between inoculum density and sugarbeet variety for stand and many harvest parameters. Moderate and susceptible varieties had more severe disease, and disease developed at lower inoculum densities. Sugarbeet growers who choose a variety with a high level of resistance to *R. solani* can expect less RCRR and less reduction in stand, yield, and recoverable sugar. Alternatively, growers that choose a susceptible variety will need a full-season control strategy including an at-planting (seed or in-furrow) and postemergence fungicide.

## ACKNOWLEDGEMENTS

We thank the Sugarbeet Research and Education Board of Minnesota and North Dakota for funding this research; Betaseed and Hilleshog for providing seed; the University of Minnesota, NWROC, Crookston for providing land, equipment, and other facilities; Jeff Nielsen for plot maintenance; Katie Baird, and Chris Larson for technical assistance; and American Crystal Sugar Company, East Grand Forks, MN for quality analysis.

## LITERATURE CITED

1. Bolton, M.D., L. Panella, L. Campbell, and M.F.R. Khan. 2010. Temperature, moisture, and fungicide effects in managing Rhizoctonia root and crown rot of sugarbeet. *Phytopathology* 100:689-697.
2. Brantner, J.R. 2013. Effect of Rhizoctonia solani inoculum density and sugarbeet variety susceptibility on disease onset and development. 2012 Sugarbeet Res. Ext. Rept. 43:236-240.
3. Brantner, J.R. and C.E. Windels. 2011. Efficacy of in-furrow and postemergence fungicides in controlling Rhizoctonia on sugarbeet. 2010 Sugarbeet Res. Ext. Rept. 41:246-249.
4. Culbreath, A.K., M.K. Beute, and C.L. Campbell. 1991. Spatial and temporal aspects of epidemics of Cylindrocladium black rot in resistant and susceptible peanut genotypes. *Phytopathology* 81:144-150.
5. Jacobsen, B.J., N.K. Zidack, M. Johnston, A.T. Dyer, K. Kephart, and J. Ansley. 2005. Studies on optimal timing of azoxystrobin applications for Rhizoctonia crown and root rot control. 2004 Sugarbeet Res. Ext. Rept. 36:291-294.
6. Khan, M.F.R., C.A. Bradley, J. Khan, and R. Nelson. 2004. Efficacy of Quadris on control of Rhizoctonia root and crown rot in 2003. 2003 Sugarbeet Res. Ext. Rept. 34:250-251.
7. Khan, M.F.R., R. Nelson, C.A. Bradley, and J. Khan. 2005. Developing a management strategy for controlling Rhizoctonia root and crown rot in sugarbeet. 2004 Sugarbeet Res. Ext. Rept. 36:295-296.
8. Kirk, W.W., P.S. Wharton, R.L. Schafer, P. Tumbalam, S. Poindexter, C. Guza, R. Fogg, T. Schlatter, J. Stewart, L. Hubbell, and D. Ruppel. 2008. Optimizing fungicide timing for the control of Rhizoctonia crown and root rot of sugar beet using soil temperature and plant growth stages. *Plant Dis.* 92:1091-1098.
9. Navas-Cortes, J.A., A. Alcalá-Jimenez, B. Hau, and R.M. Jimenez-Diaz. 2000. Influence of inoculum density of races 0 and 5 of *Fusarium oxysporum* f. sp. *ciceris* on development of Fusarium wilt in chickpea cultivars. *European J. Plant Pathol.* 106:135-146.
10. Niehaus, W.S. 2012. Results of American Crystal's 2011 official coded variety trials. 2011 Sugarbeet Res. Ext. Rept. 42:276-346.
11. Windels, C.E. and J.R. Brantner. 2005. Early-season application of azoxystrobin to sugarbeet for control of *Rhizoctonia solani* AG 4 and AG 2-2. *J. Sugar Beet Res.* 42:1-17.
12. Xiao, C.L. and K.V. Subbarao. 1998. Relationships between *Verticillium dahliae* inoculum density and wilt incidence, severity, and growth of cauliflower. *Phytopathology* 88:1108-1115.