Northern NY Agricultural Development Program
2012 Project Report

Alfalfa Germplasm from the Presence of the Brown Root Rot Fungus and Ice Sheeting

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Background:

Phoma sclerotioides, causal agent of brown root rot (BRR), is a soil-borne fungus causing root and crown rot of alfalfa, other perennial legumes, and overwintering grasses. Primarily active during late winter and early spring (Cormack 1934), it is associated with yield loss, winterkill, slow emergence from winter dormancy, and stand decline of alfalfa (Berkenkamp et al. 1991, Hollingsworth et al. 2003) and with winterkill of grasses (Larsen et al. 2007).

Brown root rot was first detected in the eastern United States in 2003 in Clinton County, NY on alfalfa. The results of subsequent surveys of alfalfa production fields conducted in Clinton County in 2004 and in New York, Vermont and New Hampshire in 2005 suggest that BRR may be a serious factor impacting the health and persistence of alfalfa in the region. Brown root rot was found on a high percentage of plants in many fields, and most of the lesions caused by the disease progressed into the cortical (internal) tissues of roots and crowns (Wunsch et al. 2007). The BRR incidence observed in northeastern United States is similar to that observed in Saskatchewan, Canada, where the disease has long been recognized as a serious problem for alfalfa production.

Brown root rot can have severe effects on alfalfa yields. In Saskatchewan fields with heavy BRR disease pressure, BRR-resistant alfalfa varieties yielded 40 to 65 percent higher than BRR-susceptible varieties (second and third production years, three cuts per year); alfalfa varieties with moderate BRR resistance yielded 23 to 43 percent higher than BRR-susceptible cultivars (Berkenkamp et al. 1991).

No management tools currently exist for BRR in New York. Peace, the BRR-resistant alfalfa variety grown in Saskatchewan and Alberta, performs poorly in New York, as it is highly susceptible to other alfalfa root rots common in New York. Crop

rotation is not an effective alternative; *P. sclerotioides* produces resting structures that can persist extended periods in the soil without a suitable substrate (Cormack, 1934), it has a very broad host range, and it can survive on organic matter in the soil (Davidson, 1990).

Significant differences in BRR resistance have been observed among alfalfa varieties grown in Saskatchewan and in Wyoming (Berkenkamp et al., 1991; Hollingsworth et al., 2005). If significant differences in BRR resistance are also observed among alfalfa varieties grown in New York, adoption of the most resistant varieties by growers in fields with high BRR pressure would be expected to increase forage yields. The most resistant varieties would also serve as sources of BRR resistance for alfalfa breeding to develop varieties with higher levels of resistance in plant material that is more adapted to our region.

A field plot trial was planted at The William H. Miner Agricultural Research Institute at Chazy NY on May 4, 2009 and the trial purpose was to determine whether currently available alfalfa varieties have any resistance to BRR. It is possible that through screening of alfalfa experimental populations on fields with the BRR organism that some or many varieties may already be moderately resistant. The trial design was a split-plot with BRR inoculated and not inoculated as the main plot treatments, and alfalfa varieties as the sub-plots. Six main plots were planted. One-half of each main plot was inoculated with barley grains infected with *Phoma sclerotioides* prior to seeding. Sub-plots were 11 alfalfa varieties / experimentals. In 2010, the not inoculated plots had yields that averaged 5.47 tons per acre and the inoculated plots averaged 5.55 tons per acre. Differences among the varieties for yielding ability were significant, but inoculating with BRR had not impacted yield at this point. Plants were dug from the Vernal plots in the spring of 2010 and it was verified in Bergstrom's plant pathology lab that plants from the inoculated plots had the BRR fungus (51% incidence) in the roots and the not inoculated plots did not have the BRR fungus (1% incidence) at a significant level.

In 2011, the yields were low due to extreme wet spring conditions and extreme dry summer conditions. The not inoculated plots averaged 0.10 tons per acre dry matter more than the inoculated plots and this difference was not statistically significant (Table 1). We expected if there were BRR resistance genes in any of the varieties tested in this trial, then the resistant varieties would have similar yields in both the not inoculated and the inoculated trials.

Methods

In May 2012, it was communicated that significant ice sheeting had killed many alfalfa plants in the BRR trial and in another alfalfa trial at Chazy. The BRR trial was going into the third production year and the other alfalfa trial was going into the first production year. Thus the ice sheeting killed both older and younger alfalfa plants. In May, visual estimates of percent stand of alfalfa remaining were taken and analyzed. It was determined that the plots were damaged beyond the point where yield data from harvesting the trial would be informative. Thus a new research goal was initiated. The research goal of this project was revised to develop alfalfa populations from the surviving plants at in the BRR trial. Six varieties from the Cornell Alfalfa Breeding Program that are currently or have been recently marketed in New York were entries in the BRR trial. On June 14th, stem cuttings were taken from surviving plants in each of the six Cornell

varieties from both the inoculated and not inoculated plots. The stem cuttings were grown in the Ithaca greenhouse and seed was produced.

Results:

The average percent alfalfa stand remaining after ice-sheeting was 11 percent for both the inoculated plots and the not inoculated plots (Table 1). The inoculated plots did not have more winterkill than the not inoculated plots.

Table 1: Forage yield and percent alfalfa stand remaining in 2009 Brown Root Rot Trial at Chazy, NY after ice-sheeting event in winter 2011-2012.

	2011 -2012^ Not Inoculated				2011 -2012^ Inoculated				
Cultivar	Total 2011 Yield	2011 Yield - % Trial Mean	2012 % Stand		Total 2011 Yield	2011 Yield - % Trial Mean	2012 % Stand		2012 Average % Stand
	T/A				T/A				
Vernal	1.97	82	21	a	1.95	84	16	a	19
WL 347LH	2.36	98	16	a	2.12	92	14	a	15
Oneida Ultra	2.38	99	9		2.25	97	19	a	14
Seedway 9558	2.13	89	11		2.19	95	15	a	13
55V48	2.76	115	13	a	2.53	109	11	a	12
Guardsman II	2.43	101	15	a	2.32	101	9		12
AmeriStand 407 TQ	2.47	103	13	a	2.35	102	7		10
ReGen	2.57	107	9		2.63	114	10		9
MsSunstra 536	2.59	107	7		2.49	108	8		8
Ezra	2.49	103	6		2.35	102	9		7
N-R-Gee	2.38	99	6		2.27	98	8		7
Trial Mean	2.41		11		2.31		11		11
F-entries	7.85 **		2.09 *		2.61 **		2.10*		
LSD(.05)	0.22		9		0.34		8		
CV(%)	7.8		69.9		12.6		57.2		

^Differences between not inoculated and inoculated treatments were not statistically significant. Interactions between treatment and cultivars were not statistically significant.

From the stem cuttings taken at Chazy, seed from eight alfalfa populations were produced in the greenhouse in the fall 2012 (Table 2).

Table 2: Pedigrees of the populations produced from stem cuttings from the alfalfa plants that survived in the winterkilled Brown Root Rot Trial at Chazy in 2012.

(Guardsman II + N-R-Gee)-Chazy selection inoculated
(Guardsman II + N-R-Gee)-Chazy selection uninoculated
Seedway 9558-Chazy selection inoculated
Seedway 9558-Chazy selection uninoculated
(Ezra + Regen)-Chazy selection inoculated
(Ezra + Regen)-Chazy selection uninoculated
Oneida Ultra-Chazy selection inoculated
Oneida Ultra-Chazy selection uninoculated

It is anticipated that the selected populations may be significantly improved in resistance to BRR and may be more winter hardy. It is of interest to test these populations in a plot trial in the future.

Conclusions/Outcomes/Impacts:

If significant differences in BRR resistance are observed in one or more of the eight populations selected from the field plots at Chazy, then adoption of the most resistant varieties by growers in fields with high BRR pressure would be expected to increase forage yields. These populations were developed from alfalfa varieties developed in and adapted to New York. The most resistant varieties would also serve as sources of genes to increase the level of resistance to BRR in our alfalfa breeding program in New York.

Outreach:

Results of this research project were presented at:
-Meeting with Seedway and Allied Seed companies on February 23, 2012
-Cornell Seed Growers Field Days on July 3, 2012
-Cornell Cooperative Extension In-Service Conference on November 13, 2012

Next steps if results suggest continued work is needed in the areas of research, demonstration and/or education.

The eight populations developed will need to be tested in another plot trial, preferably on the same field that was inoculated with the BRR organism. To have enough seed for one yield trial, a second generation of plants from the seed produced on the original plants need to be grown in the greenhouse and seed produced from these plants. It is anticipated that at least 90 grams of seed will be produced (synthetic generation 2). From this trial on a field known to be infested with the BRR organism; it will be possible to determine whether the populations were improved in BRR resistance and winterhardiness. Once plants that are resistant to BRR are identified, these plants can be incorporated into new breeding lines and varieties.

Acknowledgments: Cornell University Agricultural Experiment Station

Reports/articles in which results of project have already been published.

Hansen, J. L., Viands, D. R. Forage Crops Data Summaries. *Plant Breeding Mimeo #12-3*.

Person(s) to contact for more information:

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Figure 1: Replication number 1 of the Brown root rot trial at Chazy in May 2012, after a significant ice-sheeting event killed most of the alfalfa.