# Northern NY Agricultural Development Program
## 2011 Project Report

**Project Title:** Breeding Alfalfa Varieties with Resistance to Alfalfa Snout Beetle

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**Collaborator(s):**
Chuck Burnett, Seed producer, Caldwell, ID.
Mike Hunter, Extension Educator in Jefferson County, worked with growers to identify a field site for a new trial established in spring 2008, 2009 and 2011.

**Cooperating Producers:**
Lewis County: Alfalfa snout beetles were collected along the roads just outside of Lowville, NY

Jefferson County: The alfalfa trials were planted on land prepared and owned by Doug Shelmidine, Sheland Farms in Adams, NY

**Background:**
Alfalfa snout beetle (ASB), *Otiorhychus ligustica*, is the most destructive insect pest of alfalfa in Northern New York (NNY), and is continuing to spread. Alfalfa snout beetle is currently infesting nine NNY counties and has invaded Canada across the St. Lawrence River. Otherwise, there is no other known infestation of this insect in North America.

Alfalfa snout beetle was introduced from Europe into the Port of Oswego during the middle to late 1800's in ship ballast. Alfalfa snout beetle was first discovered as a problem around 1930 after alfalfa was introduced into Oswego County. This pest causes severe yield and stand losses on alfalfa by larval feeding on alfalfa roots. New infestations are often mistaken for winter injury since the majority of plants die after the last harvest and before spring growth. With other introduced insect pests, two combined strategies have been effectively used to reduce the insect populations to manageable

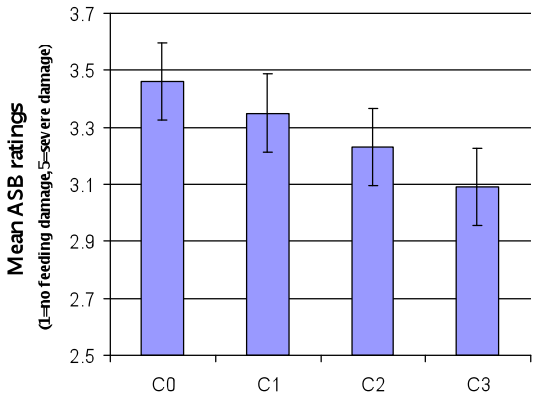
levels. These strategies are 1) identify and incorporate resistance genes into acceptable alfalfa varieties (breeding for resistance) and 2) identify and establish in NNY biological control organisms (entomopathogenic nematodes) from the native home of ASB.

None of the alfalfa varieties grown in northern USA during the 1990s appeared to be resistant when grown on a field heavily infested with ASB. In 1998 at Watertown, NY, the perennial *Medicago* core collection and other germplasms were evaluated for resistance/tolerance to root feeding damage by ASB by visually rating individual plants with a score from 1 to 5 (1 = no root damage, 5 = dead plant). The ASB damage score for 173 plant populations ranged from 3.7 to 4. This variability suggests that resistance genes may exist at a low level in a few populations. Therefore, we initiated recurrent selection to increase the level of resistance in the most resistant populations.

Because of the time-consuming and unreliable nature of field screenings, a greenhouse screening method was developed by E. J. Shields and A. Testa with funding from the NNY Agricultural Development Program. With this greenhouse screening method, the ASB population pressure can be controlled by the number of eggs applied uniformly to each flat and by the length of time that the larvae are allowed to feed on the alfalfa roots. Thus, plants with a low level of resistance can be selected and, over several cycles of selection, the frequency of resistance genes can be increased in several alfalfa populations. The first cycle of selection was completed in 11 populations in 2003 and selection has continued at the rate of one cycle per year.

An experiment was completed in the fall of 2006 under controlled greenhouse conditions to determine progress from selection. Significant progress was realized through three cycles of recurrent phenotypic selection. Averaged across alfalfa populations, root damage visually scored on a 1 (no root damage) to 5 (severe root damage) basis was 3.46 for the base populations, 3.35 for Cycle 1, 3.23 for Cycle 2, and 3.09 for Cycle 3 (see Figure 1 below). One population had a difference of more than a whole scoring unit between Cycles 0 and 3. This trend is significant and suggests that more improvements could be made by further selection. These data are the first indication that progress from selection can be made in increasing resistance to ASB.

Seed was produced in pollination cages in Idaho in 2007 in order to have enough seed of the advanced generation alfalfa selection for use in establishing plot trials in ASB-infested fields during spring 2008, 2009 and 2011 at Sheland Farms. This field research will allow comparison of Cycles 0 and 4 (as well as Cycle 6 in the 2009 trial and Cycle 8 in the 2011 trial) in three alfalfa populations to determine if the breeding efforts translate into differences in forage yield, plant stand, and root damage ratings in farmers' fields where ASB populations exist. Because of the



unreliable nature of insect infestation in field trials, trials established in two different years are necessary for conclusions about increased resistance.

| Cycle of selection and Root rating | | | |
|---|---|---|---|
| CO | C1 | C2 | C3 |
| 3.57 | 3.13 | 2.96 | 2.30 |

Figure 1: Progress in breeding alfalfa snout beetle resistant alfalfa – results from a greenhouse evaluation. Root rating was from 1 (no damage) to 5 (severe damage).

## Methods:

Breeding for Alfalfa Snout Beetle Resistance

The number of alfalfa snout beetles collected for use in the greenhouse screening procedure has been over 10,000 each year (2010 and 2011). Alfalfa seedlings were inoculated with eggs collected from the beetles. Around 35 days after inoculation, plants with the least injury were selected, interpollinated and seed produced for the next cycle of selection.

Field Evaluation of Alfalfa Snout Beetle Resistance in Alfalfa populations

The trial seeded in 2009 was harvested for yield on May 21, July 12, August 6, October 12 in 2010 and on May 25, June 30, and August 2 in 2011. The alfalfa populations seeded in the 2009 trial included three base populations, three corresponding populations after selection for four cycles of improvement for alfalfa snout beetle resistance, and two advanced cycle 6 populations. A new trial was seeded in 2011. The entries in this trial included two base populations, two corresponding populations after selection for either four, seven or eight cycles of improvement for alfalfa snout beetle resistance, and one cross of advanced germplasm.

## Results

Breeding Program for Resistance to Alfalfa Snout Beetle

In 2010 and 2011, we completed the eighth and ninth cycles of selection for resistance in 7 alfalfa populations. Plants with the least injury were selected and interpollinated within populations to produce seed for the next cycle of selection. Since 2003, a total of more than 160,000 plants have been evaluated for resistance to ASB. About 14,500 plants were evaluated in 2010 and 17,500 plants were evaluated in 2011.

Five grams of seed from the most advanced selection cycle in all seven populations has been sent to Chuck Burnett in Caldwell, ID, for increase to a few pounds of seed from each. This seed will be used to establish more field trials in coming years that will test the more advanced cycles of selection for field levels of resistance and yield.

Field Evaluation of Progress in Developing Alfalfa Snout Beetle Resistant Alfalfa
Trial #1, Sown in 2009
In the first and second production years, 'Seedway 9558' cycles 4 and 6 had significantly higher yield compared to Seedway 9558 (unselected or cycle 0)(Appendix, Table 1). 'Curculio resistant' cycle 4 had higher yield that Curculio resistant cycle 0 in the second production year. The other population, 'ASB selections' did not show significant yield improvement when comparing the advanced selections for alfalfa snout beetle resistance to the unselected populations. Between the second and third harvests in 2011, some color differences were noted among the plots. The advanced cycles of selections (cycle 6) tended to be a darker green color (rating average 2.9, 1 is light green and 3 is dark green) compared to the unselected populations (rating average 2.0). Since the alfalfa snout beetle larvae eat the alfalfa nodules where atmospheric nitrogen is fixed and is available for the alfalfa plants, the lighter green color may be an initial indication of more severe root feeding damage. At this same time, height of the plots was also measured, but differences were not associated with selection cycle. The average yield in 2011 of the unselected or cycle 0 populations was 4.11 tons per acre, of the cycle 4 populations was 4.33 tons per acre and of the cycle 6 populations was 4.70 tons per acre (Appendix, Figure 1).

Trial #2, Sown in 2011 The trial sown in 2011 was managed for good alfalfa establishment, but was not harvested for yield. This trial will be harvested in 2012 and beyond. Plant stands in the seeding year were excellent.

## Conclusions/Outcomes/Impacts:

In both the greenhouse evaluation and field evaluation of the breeding progress made in alfalfa resistance to the alfalfa snout beetle, positive results showed that the selection program is successful and can be used to find alfalfa seedlings that have some level of tolerance or resistance to root feeding damage. For these two evaluations, a limited number of populations and a limited number of cycles of selection were tested. From the 2012 NNYADP grant, we will have the seed to test more populations and advanced lines that have been in the breeding program that began in 2002 that have not been evaluated yet.

It is anticipated that the first alfalfa snout beetle resistant alfalfa variety, the Seedway 9558 cycle 7 population or NY1010, will be named and seed will be available for sale in 2014. Producers in the ASB infested areas of NY are eager to plant an alfalfa variety with tolerance / resistance to the beetle. Producers will need to be aware that an alfalfa variety with strong resistance to ASB may not be available until more cycles of selection are completed and more crosses are made.

## Outreach:

Results of this research project were presented at:
-Meeting with Seedway and Allied Seed companies on January 13, 2010 and March 8, 2011
-Cornell Seed Growers Field Days on July 8, 2010 and July 7, 2011
-Cornell Cooperative Extension In-Service Conference on November 17, 2010 and November 16, 2011
-Jefferson County Field Day on August 2, 2011 at Sheland Farms
-New article by Kara Dunn in August, 2011
- Powerpoint presentation about the alfalfa snout beetle resistant alfalfa to grower's interested in raising their own nematodes on March 6, 14, and 15 in 2012 in Northern New York

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

The breeding program for resistance needs to continue so that more cycles of selection can be accomplished and more populations screened. Genetic improvement in alfalfa is accomplished by step-wise accumulation of numerous favorable genes. We expect as selection continues, that the level of resistance to alfalfa snout beetle will increase. Along with the breeding program, evaluation of the alfalfa improvement progress will be important to document. Larger field scale strip trial will be possible in the future once commercial quantities of seed are available.


## Acknowledgments:

CUAES Hatch Funds, NE1010 Regional Research Funds, Seedway and Allied LLC.


## Reports and/or articles in which the results of this project have already been published.

Two-Pronged Attack Thwarts Snout Beetles, Hay and Forage Grower, September 20, 2011

Research shows promise for controlling destructive alfalfa snout beetle, Cornell Chronicle, September 14, 2011

## Person(s) to contact for more information (including farmers who have participated: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**Appendix**:

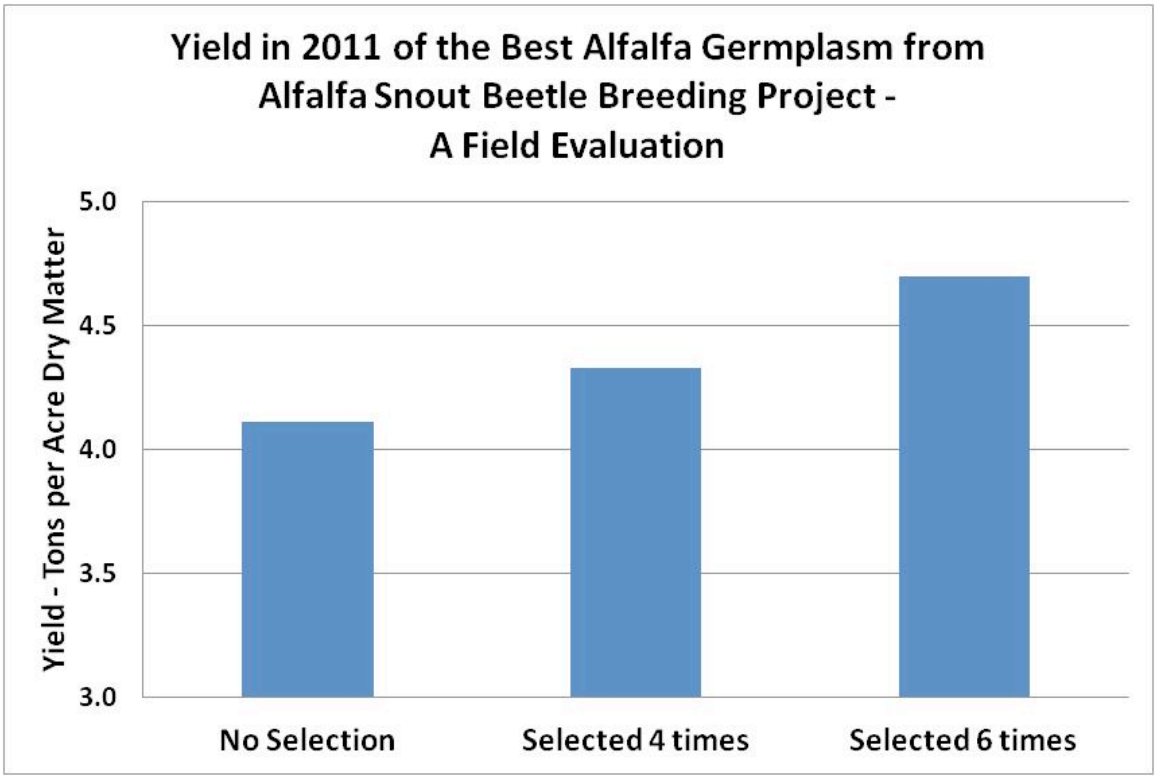


Figure 1: Second production year yields of alfalfa cultivars and populations selected for resistance to alfalfa snout beetle either zero time, four times, or six times.