**M.L. 2010 Project Abstract**
For the Period Ending June 30, 2014

**PROJECT TITLE:** Biological Control of European Buckthorn and Garlic Mustard
**PROJECT MANAGER:** Laura Van Riper
**AFFILIATION:** Minnesota Department of Natural Resources
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**FUNDING SOURCE**: Environment and Natural Resources Trust Fund
**LEGAL CITATION:** M.L. 2010, Chp. 362, Sec. 362, Subd. 6a and M.L. 2013, Chapter 52, Section 2, Subdivision 17

**APPROPRIATION AMOUNT: $300,000**

## Overall Project Outcome and Results

European/common buckthorn (*Rhamnus cathartica*) and garlic mustard (*Alliaria petiolata*) are non-native invasive plants that severely threaten native plant communities and degrade wildlife habitat. They are widely distributed in the state and current control options, such as mechanical and chemical control, are labor and cost-intensive. They are of the highest priority for development of long-term management solutions, such as biological control. The purpose of this research was to determine 1) if there are suitable insects that can be used to reduce impacts caused by buckthorn and 2) implement introduction of insects to control garlic mustard and assess their establishment and success.

Over 30 specialized insects were identified as potential common buckthorn biocontrol. Most of these species were discarded because they lacked host-specificity. Two psyllids were host-specific, but did not cause significant damage to buckthorn and the insects were infected with the plant disease '*Candidatus* Phytoplasma rhamni' (buckthorn witches' broom). A seed-feeding midge proved too difficult to work with in a research setting. After 11 years of searching for a biological control insect that is host-specific and damaging to buckthorn, we conclude that there are not promising agents at this time.

Four *Ceutorhynchus* weevil species are being studied as biological control agents for garlic mustard. Petitions for release were submitted to the USDA-APHIS Technical Advisory Group starting in 2008, but they have requested additional host-specificity testing over time. No biological control insects have been approved for release as of 2014. Studies conducted in the University of Minnesota Containment Facility allowed the development of efficient and consistently reliable methods to rear *C. scrobicollis* from garlic mustard plants. Long-term monitoring at twelve sites in Minnesota shows that garlic mustard populations can fluctuate widely from year to year. There is little garlic mustard herbivory in Minnesota. Garlic mustard cover is negatively correlated with cover of other species.

## Project Results Use and Dissemination

**Buckthorn biological control research has been disseminated in the following ways:**
*Peer reviewed journal publication (pdf attached):*

- Gassmann, A. and I. Tosevski. 2014. Biological control of Rhamnus cathartica: is it feasible? A review of work done in 2002–2012. Journal of Applied Entomology 138: 1-13.

*CABI Report Summary (pdf attached):*

- Gassmann, A., A. Leroux, M. Bennett, M. Penic, N. Haefliger, R. Eschen, J. Jović and I. Toševski. 2012. Report 2010–12: Biological control of common buckthorn, *Rhamnus cathartica*. CABI Europe-Switzerland. CABI Ref: VM01730.

*Poster presentations at conferences:*

- Gassman, Andre, Laura C. Van Riper*, and Luke C. Skinner. Conclusions from 11 Years of Buckthorn Biocontrol Research. Ecological Society of America Conference. 4-9 August 2013. La Crosse, WI.
- Gassman, Andre, Laura C. Van Riper*, and Luke C. Skinner. Conclusions from 11 Years of Buckthorn Biocontrol Research. Upper Midwest Invasive Species Conference. 29-31 Oct 2012. La Crosse, WI.
- Gassman, Andre, Laura Van Riper*, and Luke C. Skinner. Developing Biological Control for Common and Glossy Buckthorn. Invasive Plants Symposium, Dec. 2011. Milwaukee, WI.
- Gassman, Andre, Laura Van Riper*, and Luke C. Skinner. Developing Biological Control for Common and Glossy Buckthorn. Minnesota-Wisconsin Invasive Species Conference, 2-10 Nov 2010. St. Paul, MN.

*Webpage created on MN DNR website:*

- http://www.dnr.state.mn.us/invasives/terrestrialplants/woody/buckthorn/biocontrol.html

**Garlic mustard biological control research has been disseminated in the following ways:**
*Peer reviewed journal publication (pdf attached):*

- Becker, R.L., E.J.S. Katovich, H.L. Hinz, E. Gerber, D.W. Ragsdale, R.C. Venette, D.N. McDougall, R. Reardon, L.C. Van Riper, L.C. Skinner, and D.A. Landis. 2013. The Garlic Mustard (*Alliaria petiolata*) Case, What Makes a Good Biological Control Target. The Intersection of Science, Perspectives, Policy and Regulation. pp. 332-339 *In* Proc. XIII International Symposium on Biological Control of Weeds (ISBCW). Sept. 11-16, 2011. Waikoloa, Hawaii. Wu, Yun; Johnson, Tracy; Sing, Sharlene; Raghu, S.; Wheeler, Greg; Pratt, Paul; Warner, Keith; Center, Ted; Goolsby, John; and Reardon, Richard, Editors. USDA Forest Service, FHTET-2012-07. January 2013. 536 p. http://www.invasive.org/publications/xiiisymposium/

*U.S. Forest Service Technology Transfer document:*

- Becker, Roger, Esther Gerber, Hariet L. Hinz, Elizabeth Katovich, Brendon Panke, Richard Reardon, Mark Renz, and Laura Van Riper. 2013. Biology and Biological Control of garlic Mustard. US Forest Service Forest Technology Enterprise Team publication FHTET-2012-05. http://www.fs.fed.us/foresthealth/technology/pdfs/GarlicMustardBiocontrol_FHTET-2012-05.pdf [Accessed May 2014].

*Reports to the Environment and Natural Resources Trust Fund (pdfs attached):*

- Katovich, E.J. and Becker, R.L. 2014. Garlic mustard biological control: Developing biological control insects, working towards field release.
- Van Riper, L.C. and Becker, R.L. 2014. Garlic mustard (*Alliaria petiolata*) monitoring in Minnesota: 2005-2013.

*Presentations:*

- E. J. S. Katovich. Upper Midwest Invasive Species Conference. November, 2010. St. Paul, MN. Biocontrol of Garlic Mustard and Buckthorn, an Update.

- E. J. S. Katovich. XIII International Symposium on Biological Control of Weeds. September, 2011. Waikoloa, Hawaii. Biological Control of Garlic Mustard, *Alliaria petiolata*, with the Root and Crown- Boring Weevil, *Ceutorhynchus scrobicollis*.
- E. J. S. Katovich. Ontario Invasive Plant Council, Annual General Meeting and Conference. October, 2011. Picton, Ontario. Potential for the Biological Control of Garlic Mustard.
- E. J. S. Katovich. Upper Midwest Invasive Species Conference. October, 2012. La Crosse, WI. Biological Control of Garlic Mustard with a Seed-Feeding Weevil.
- E. J. S. Katovich. Biological Control of Northeastern Weeds-2013 Cooperators Meeting. February, 2013. Trenton, New Jersey. Garlic Mustard Biocontrol: Current Status and Future Directions.
- E. J. S. Katovich. Technical Advisory Group For the Biological Control of Weeds, Annual Meeting. June, 2013. Washington, D.C. *Ceutorhynchus scrobicollis* as a Potential Biocontrol Agent of Garlic Mustard, *Alliaria Petiolata.*
- E. J. S. Katovich. University of Minnesota, guest lecturer for AGRO 4505: Biology, Ecology and Management of Invasive Plants. Biological Control of Invasive Plants. 2010-2014.

# 2010 Environmental and Natural Resources Trust Fund ENRTF
# Work Program Amendment Final Report

**Date of Report:** August 15, 2014
**Date of Next Status Report:** Final Report
**Date of Work program Approval:** June 3, 2010
**Project Completion Date:** June 30, 2014

**I. PROJECT TITLE: Biological Control of European Buckthorn and Garlic Mustard**

**Project Manager**: Laura Van Riper
**Affiliation:** Minnesota Department of Natural Resources
**Address:** 500 Lafayette Road, Box 25
St. Paul, MN 55155-4025
**Telephone number**: 651-259-5090
**Email**: laura.vanriper@state.mn.us

**Location:** State, county and federal parks, forests, nature preserves and wildlife management areas; roadsides private woodlots and agricultural lands statewide.

**Total ENRTF Project Budget:** **ENRTF Appropriation: $ 300,000**
**Minus Amount Spent: $ 300,000**
**Equal Balance: $ 0**

**Legal Citation: M.L. 2010, Chp. 362, Sec. 362, Subd. 6a** and **M.L. 2013, Chapter 52, Section 2, Subdivision 17**

**Appropriation Language:**
The availability of the appropriations for the following projects are extended to June 30, 2014: (6) Laws 2010, chapter 362, section 2, subdivision 6, paragraph (a), Biological Control of European Buckthorn and Garlic Mustard

$300,000 is from the trust fund to the commissioner of natural resources in cooperation with the commissioner of agriculture to continue the development and implementation of biological control for European buckthorn and garlic mustard. This appropriation 14.2 is available until June 30, 2014, by which time the project must be completed and final products delivered.

An extension is requested to complete activity 2: Introduction and evaluation of Garlic Mustard biological control agents in MN. While a petition for release of the biocontrol weevil *Ceutorhynchus scrobicollis* was submitted to the USDA in 2011, no decision has been made by the agency. Without the insect being approved for release, we were not able to complete the tasks in the work plan. We are requesting a one year extension to continue to work on the actions in Activity 2. This one-year extension is contingent on legislative approval.

Amendment Approved: May 9, 2013

## II. FINAL PROJECT SUMMARY AND RESULTS:

European/common buckthorn (*Rhamnus cathartica*) and garlic mustard (*Alliaria petiolata*) are non-native invasive plants that severely threaten native plant communities and degrade wildlife habitat. They are widely distributed in the state and current control options, such as mechanical and chemical control, are labor and cost-intensive. They are of the highest priority for development of long-term management solutions, such as biological control. The purpose of this research was to determine 1) if there are suitable insects that can be used to reduce impacts caused by buckthorn and 2) implement introduction of insects to control garlic mustard and assess their establishment and success.

Over 30 specialized insects were identified as potential common buckthorn biocontrol. Most of these species were discarded because they lacked host-specificity. Two psyllids were host-specific, but did not cause significant damage to buckthorn and the insects were infected with the plant disease '*Candidatus* Phytoplasma rhamni' (buckthorn witches' broom). A seed-feeding midge proved too difficult to work with in a research setting. After 11 years of searching for a biological control insect that is host-specific and damaging to buckthorn, we conclude that there are not promising agents at this time.

Four *Ceutorhynchus* weevil species are being studied as biological control agents for garlic mustard. Petitions for release were submitted to the USDA-APHIS Technical Advisory Group starting in 2008, but they have requested additional host-specificity testing over time. No biological control insects have been approved for release as of 2014. Studies conducted in the University of Minnesota Containment Facility allowed the development of efficient and consistently reliable methods to rear *C. scrobicollis* from garlic mustard plants. Long-term monitoring at twelve sites in Minnesota shows that garlic mustard populations can fluctuate widely from year to year. There is little garlic mustard herbivory in Minnesota. Garlic mustard cover is negatively correlated with cover of other species.

## III. PROGRESS SUMMARY AS OF *(12/30/13)*:

**Update (12/30/13):**

The buckthorn project (Activity 1) had been completed as of 6/30/2013. Since that time a journal article resulting from that work has been accepted for publication by the Journal of Applied Entomology.

A contract with the University of Minnesota for the garlic mustard work in Activity 2 was written and signed. Garlic mustard monitoring data was collected in October 2013. Also in October 2013 a letter was submitted to the USDA APHIS Technical Advisory Group (TAG) with the plant species that we propose for additional host-specificity testing.

**Update (06/30/13):**

Buckthorn biocontrol research was completed and a final report submitted by CABI. CABI has also developed a draft journal article that will be submitted for publication in the journal "Biological Control". The final report is attached to this summary. Numerous potential biocontrol insects for common and glossy buckthorn were screened for host-specificity and impacts. After 11 years of searching for a biocontrol insect that is both host-specific and damaging to common buckthorn, we conclude that we do not have any promising agents at this time. The journal article will be summarize the results of the buckthorn biocontrol research so the results will be available if a new buckthorn biocontrol project is initiated in the future.

Garlic mustard monitoring of field sites was conducted in October 2012 and June 2013. Lab studies continued to develop mustard propagation methods and *C. scrobicollis* rearing protocols in the High Containment facility the University of Minnesota in anticipation of permission to release *C .scrobicollis* into the field for the biocontrol of garlic mustard. In May and June 2013 we received communication from the USDA APHIS Technical Advisory Group regarding the petition for release of *C. scrobicollis*. The petition was rejected based on the desire to see additional Threatened and Endangered mustard species undergo host-specificity testing. Dr. Katovich and Dr. Becker will work with the US Fish and Wildlife Service on developing a list of species to be tested and obtaining the seeds. At this time, we do not know the final number of additional species that need to be tested, but it appears that it will be fairly small as many of the species have already been tested.

**Update (09/30/12):**
Buckthorn biocontrol research was completed and a final report submitted by CABI. Due to the difficulties surrounding currently studied agents and the low probability of finding additional potential agents, it has been decided that buckthorn biocontrol research will not be pursued into the future. The remaining psyllid potential biocontrol agents had issues with a lack of impact on buckthorn and potentially carrying a phytoplasma (plant disease) that is not known to be in the United States. The researchers were not able to work with the remaining seed-feeding midge in a research setting as they could not obtain fruiting trees of buckthorn species. The research that has been done on buckthorn biocontrol will be written into a journal article so that others may learn from this research and to provide a starting point if someone were to reinitiate buckthorn biocontrol research in the future.

There has been no notification from the USDA Technical Advisory Group (TAG) as to the status of the petition for the release of *Ceutorhynchus scrobicollis* as a biological control insect for garlic mustard. The petition was submitted to TAG in September of 2011. Garlic mustard monitoring data was collected from the 12 permanent monitoring sites in June 2012. Studies to maximize the reliability and production of *C. scrobicollis* rearing have been carried out. A 3 month aestivation time allows for the greatest production of insects. A study looking at the effect of the soil mixtures has found that the addition of 3 to 4 cm of greenhouse soil can aid in *C. scrobicollis* pupal survival and result in an increase in the number of insects reared. Work continues on updating and revising a manual for propagating garlic mustard and rearing *C. scrobicollis*.

**Update (02/28/12):**
Garlic mustard monitoring was conducted in October 2011. Data has been entered and data analysis has begun. Garlic mustard continues to be widespread. Of the 12 monitoring sites, the average percent cover of garlic mustard in June 2011 ranged from 6% garlic mustard cover at the lowest cover site to 65% garlic mustard cover at the highest cover site. There has been no notification from the USDA Technical Advisory Group (TAG) as to the status of the petition for the release of *Ceutorhynchus scrobicollis* as a biological control insect for garlic mustard.

Buckthorn biological control research at CABI Europe-Switzerland found that the potential biocontrol insect *Trichochermes walkeri* proved to be infected with '*Ca.* Phytoplasma rhamni' at a very high rate in almost all sampled localities. In Europe, *R. cathartica* trees were found to be infected with '*Candidatus* Phytoplasma rhamni' at almost all surveyed localities. Researchers did not find evidence of negative plant-soil feedback by mature *R. cathartica* on conspecifics that could explain low seedling numbers of *R. cathartica* in the native range.

Sixty buckthorn plants throughout Minnesota and the Midwest were sampled and tested for the presence of the buckthorn phytoplasma disease '*Candidatus* Phytoplasma rhamni'. None of the samples were found to be positive for the phytoplasma.

**Update (09/30/11):**
The garlic mustard biocontrol host-specificity was completed by researchers at CABI Europe-Switzerland and the University of Minnesota. The results were written up and the petition was submitted to the USDA Technical Advisory Group (TAG) on September 8, 2011. The petition was a supplement to the original petition number 08-05, submitted April 2008. The petition title was: A Petition for the Introduction, Experimental Release and Open-Field Release of the Root-Mining Weevil *Ceutorhynchus scrobicollis* (Coleoptera: Curculionidae) for the Biological control of Garlic Mustard (*Alliaria petiolota*) in North America . The petitioner was Dr. Luke Skinner. The Technical Advisory Group will review the petition and recommend to USDA-APHIS whether or not the garlic mustard biocontrol insect *C. scrobicollis* should be approved for release in the United States.

Research continues at CABI Europe-Switzerland on buckthorn biological control. Work focuses on the insects *T. walkeri, W. krumbholzi* and the phytoplasma disease '*Candidatus* Phytoplasma rhamni'. Using LCCMR funds, a contract was written with Dr. Roger Becker at the University of Minnesota to test buckthorn plants from the United States for presence of the phytoplasma.

Using LCCMR funds, a contract was written with Dr. Roger Becker at the University of Minnesota for garlic mustard monitoring in June and October 2011.

**Update (02/28/11):** A two year contract was written with CABI Europe-Switzerland for continued research on buckthorn biological control. Goals for July 1, 2010 to June 30, 2011 include continuing to assess the feasibility of using insects *Trichochermes walkeri*, *Cacopsylla rhamnicolla*, and *Wachtiella krumbholzi* as biological control agents for *Rhamnus cathartica*. Additional study of the plant disease phytoplasma '*Candidatus* Phytoplasma rhamni' is necessary to determine if *T. walkeri* could be used as a biological control agent. Additionally, researchers will work to determine the causes of the high levels of seed and seedling mortality of *R. cathartica* observed in Europe as a step toward identifying additional potential biological control agents. CABI researchers have collected samples of *Rhamnus* species and *T. walkeri* for detection of the phytoplasma. Samples of the *Rhamnus* have been analyzed and the phytoplasma has been detected in four of the countries they sampled, but trees did not show visible symptoms of the disease. Additional work has been completed in preparation for additional host specificity testing of the target insects.

A contract was written with the University of Minnesota for garlic mustard monitoring in October 2010. The results of that research are currently being analyzed. Final host specificity testing of the garlic mustard biocontrol agent *Ceutorhynchus scrobicollis* is expected to be completed by May 2011. Then a proposal for approval for release will be submitted to the USDA-APHIS Technical Advisory Group. TAG will give a recommendation as to whether the insects may be released in the United States.

## IV. OUTLINE OF PROJECT RESULTS*:*

**Result/Activity 1:** Investigate potential insects as biological control of European Buckthorn

**Description:** Researchers from the CABI Europe-Switzerland will continue to locate, identify and collect potential natural enemies of *Rhamnus cathartica* and *Frangula alnus* of *Rhamnus* spp in Europe. Host specificity studies (make sure the insects will not eat plants native to MN and the U.S.) will continue on the high priority insect species. Insects will be prioritized based on their perceived potential to cause damage to buckthorn by impairing growth and/or reproduction, reduce vigor, or cause structural damage. These factors can potentially lead to buckthorn mortality. Expected results include a priority list of potential control agents with information on their host specificity to native buckthorn species and other plants as determined. This information will guide future research and eliminate candidate insects that are not good potential agents. Testing is done in Europe due to availability if insects and reduce risk of importing any species prior to release. Most species are collected from the wild as cuttings or as seed. Precautions are taken to ensure no soil or other plant parts are shipped with the test plants. The plants are then grown by the researcher in Switzerland and used in testing the insects. Testing procedures are determined once the insects have been identified.

| **Summary Budget Information for Result/Activity 1:** | **ENRTF Budget** | **$150,000** |
|---|---|---|
| | **Amount Spent** | **$150,000** |
| | **Balance** | **$0** |

| Deliverable/Outcome | Completion Date | Budget |
|---|---|---|
| Field collection and host specificity testing of agents in 2010 and annual report summarizing results for 2010 | 2/28/11 | $30,000 |
| Field collection and host specificity testing of agents in 2011 | 9/30/11 | $30,000 |
| Annual report summarizing results for 2011 | 2/28/12 | $30,000 |
| Field collection and host specificity testing of agents in 2012 | 9/30/12 | $30,000 |
| Final report with findings and recommendations | 6/30/13 | $30,000 |

**Completion Date: 6/30/14**

**Results Status as of (12/30/13):**
This project was completed as of 6/30/2013. The research paper that resulted from this work was accepted for publication by Journal of Applied Entomology. The article is currently in press.

**Results Status as of (6/30/13):**
Buckthorn biocontrol research was completed and a final report submitted by CABI (Attached). CABI has also developed a draft journal article that will be submitted for publication in the journal "Biological Control". Numerous potential biocontrol insects for common and glossy buckthorn were screened for host-specificity and impacts. Early on, glossy buckthorn biocontrol was eliminated from consideration due to lack of promising agents. Research continued on common buckthorn. After 11 years of searching for a biocontrol insect that is both host-specific and damaging to common buckthorn, we conclude that we do not have any promising agents at this time. The journal article will summarize the results of the buckthorn biocontrol research so the results will be available if a new buckthorn biocontrol project is initiated in the future.

**Result Status as of (9/30/12)**
**Research in Europe:** An impact study of the effect of leaf galling by *T. walkeri* on eight-month-old *R. cathartica* seedlings was set up in August 2011. A total of 714 eggs were laid on infected trees. However, in 2012, no galls were recorded and the test was terminated without having obtained conclusive results.

Buckthorn biocontrol research projects were completed; work now focuses on writing the results and conclusions. CABI submitted their final report to MN DNR in September 2012. There is low potential for the remaining potential biocontrol insects to provide control of buckthorn. The psyllid species may be implicated in the spread of the phytoplasma (a type of plant disease) witches'-broom of buckthorn. Many buckthorn plants in Europe have the phytoplasma, but show no symptoms. There is no evidence that the buckthorn phytoplasma is present in the US. This possibility of a biocontrol agent spreading a phytoplasma that is not already present in the US makes it unlikely that the insects would gain approval for release. The remaining biocontrol insects, the seed feeding midges, proved too difficult to work with in a lab setting. The researchers could not obtain reproductive trees of buckthorn species, so therefore could not pollinate female buckthorn flowers or synchronize fruit development with midge oviposition and larval development. Without fruits of buckthorn species, the researchers could not screen the midges as to their host specificity.

Due to the difficulties surrounding currently studied agents and the low probability of finding additional potential agents, it has been decided that buckthorn biocontrol research will not be pursued into the future. The lead researcher will write up the final results of the buckthorn biocontrol research project for publication.

**Result Status as of (2/28/12)**
**Research in Europe:**

- In Europe, *R. cathartica* trees were found to be infected with '*Candidatus* Phytoplasma rhamni' at almost all surveyed localities, although the presence of witches' broom symptoms were not observed. Phytoplasma was not detected in any of the other *Rhamnus* species analyzed, which suggests a very specific host association of this phytoplasma with its plant host.
- *Trichochermes walkeri* proved to be infected with '*Ca.* Phytoplasma rhamni' at a very high rate in almost all sampled localities. However, *T. walkeri* infection with phytoplasma only shows that this psyllid is acquiring the phytoplasma during feeding on infected plants, but not a capability to re-inject the phytoplasma during feeding. The latter will be tested in transmission trials that were started in 2011 and will be completed in 2012.
- Researchers did not find evidence of negative plant-soil feedback by mature *R. cathartica* on conspecifics that could explain low seedling numbers of *R. cathartica* in the native range.

**Research in the US:**
Sixty buckthorn plants throughout Minnesota and the Midwest were sampled and tested for the presence of the buckthorn phytoplasma disease '*Candidatus* Phytoplasma rhamni'. None of the samples were found to be positive for the phytoplasma.

**Result Status as of (9/30/11)**
**Research in Europe:**

- 100 *T.walkeri* adults were collected in Serbia from *Rhamnus cathartica* trees which had known to be infected with '*Ca.* Phytoplasma rhamni'. A phytoplasma transmission experiment was set up to see if those insects can infect non-infected trees.
- A study was initiated on the potential impact of *T. walkeri* on young buckthorn plants.

- About 30 adults of *W. krumbholzi* were collected, but none of the lab's potted *R. cathartica* flowered or fruited, thus no oviposition tests could be carried out. Researchers suggest discarding *W. krumbholzi* from the list of potential agents.
- An experiment was established to test the hypothesis that seed and seedling mortality of *R. cathartica* in Europe is affected by negative plant–soil feedbacks.

**Research in the US:** The presence of the phytoplasma in the potential biocontrol agent *T. walkeri* raises questions about the possibility of using this agent for biocontrol. It is not known if the phytoplasma is present in North American populations of buckthorn, but this is necessary information to assess the potential for *T. walkeri* as a biocontrol agent. Researchers at the University of Minnesota conducted a preliminary survey for this phytoplasma in Minnesota and coordinated with partners in other states to have them send in buckthorn samples for laboratory analysis. The results of the lab testing are not available at this time.

**Result Status as of (02/28/11):** The 2010 annual report will be submitted March 2011. '*Candidatus* Phytoplasma *rhamni*' is a witches'-broom disease of European buckthorn (*Rhamnus cathartica*). Phytoplasmas are non-culturable, insect-transmitted, wall-less bacteria. In 2009, the presence of '*Candidatus* Phytoplasma *rhamni*' was detected in the adults of the potential biocontrol insect *Trichochermes walkeri* (a sap-sucking psyllid) in two locations in Switzerland. Psyllids may be vector for transmitting the virus. The biology and transmittal of the phytoplasma is not well understood. Additional research on the phytoplasma and *T. walkeri* is necessary to assess whether *T. walkeri* is still viable as a potential biocontrol insect for buckthorn in the United States.

In 2010, CABI researchers sampled trees (*R. cathartica*, other *Rhamnus* species, *Frangula alnus*) and insects (*T. walkeri*) in a number of sites within five countries in Western Europe for the detection of the phytoplasma '*Candidatus* Phytoplasma *rhamni*'. The phytoplasma was detected in *R. cathartica* samples at several sites in all countries surveyed, except for Montenegro, but not in any of the other *Rhamnus* species sampled or in *F. alnus*. The researchers did not observe symptoms of the witches'-broom disease and cannot associate the presence of the phytoplasma with any particular symptoms in the trees. A high rate of phytoplasma has been found at the two sites where the positive *T. walkeri* samples had been collected in 2009. The psyllid *T. walkeri* samples collected in 2010 are being analyzed and results will be available by the end of March 2011.

In preparation for continued host specificity testing of the gall midge *Wachtiella krumbholzi*, CABI researchers collected buckthorn fruits attacked by the gall midge. They can use these insects for host-specificity testing. Additionally, CABI researchers collected mature fruits and seeds of *R. cathartica* in Europe to be used in a plant-soil feedback study. This study may identify other potential biocontrol agents and help explain why buckthorn is held in check in Europe. They have also done germination experiments in order to prepare for this soil feedback study.

**Final Report Summary:**

Biological control of common buckthorn (*Rhamnus cathartica*) research focused on assessing the feasibility of using the psyllids *Trichochermes walker*, *Cacopsylla rhamnicola*, and *Trioza rhamni* and the seed-feeding midge *Wachtiella krumbholzi* as biological control agents, determining the biology and transmittal of the witches broom disease '*Candidatus* Phytoplasma rhamni' which

was found to be present in the psyllids, and determining the causes of the high levels of seedling mortality and post-dispersal seed mortality of common buckthorn observed in Europe as compared to North America.

While research indicated the three psyllid species were host-specific to common buckthorn, there were two issues that complicated their use as biocontrol insects. There was the potential that the psyllids could bring the buckthorn witches broom disease ('*Ca.* Phytoplasma rhamni') to the United States. It was also not clear that the psyllids could cause enough damage to common buckthorn to be an effective control agent.

Little was known about 'Ca. Phytoplasma rhamni' so additional research was necessary. In Europe, common buckthorn trees were found to be infected with '*Ca.* Phytoplasma rhamni' at almost all surveyed localities, confirming previous reports of host association of this phytoplasma with common buckthorn, although the presence of witches' broom symptoms were not observed. The phytoplasma was not detected in any of the other *Rhamnus* species analyzed, which suggests a very specific host association of this phytoplasma with its plant host, and also a very specific relationship between the insect vector of the pathogen and its host plant. Work on '*Ca.* Phytoplasma rhamni' in North America was carried out by Dr. Roger Becker and Dr. Dimitre Mollov, University of Minnesota, St Paul, USA. '*Ca.* Phytoplasma rhamni' was not detected in 75 *R. cathartica* populations from North America suggesting either that the phytoplasma has not been introduced in the exotic range of its host plant, or that the absence of a suitable vector for phytoplasma propagation constrained its establishment in North America. *Trichochermes walkeri* proved to be infected with '*Ca.* Phytoplasma rhamni' at a very high rate in almost all sampled localities. Transmission trials strongly suggest that *T. walkeri* is not a vector of '*Ca.* Phytoplasma rhamni'. *Trichochermes walkeri* acquires the phytoplasma during feeding on infected plants, but it is not capable of re-injecting the phytoplasma during feeding. The phytoplasma was also found in *Cacopsylla rhamnicola* and *Trioza rhamni* although the role they play in spreading the phytoplasma is not clear.

An impact study of the effect of leaf galling by *T. walkeri* on eight-month-old *R. cathartica* seedlings was set up in August 2011. A total of 714 eggs were laid on infected trees. However, in 2012, no galls were recorded and the test was terminated without having obtained conclusive results.

The seed-feeding midge *W. krumbholzi* was found at most common buckthorn (*R. cathartica*) sites where searched. Midge larvae have also been discovered in the fruits of rock buckthorn (*R. saxatilis* ssp. *tinctorius*) at one site in Serbia, where common buckthorn also occurs. Based on the mitochondrial COI (cytochrome c oxidase) gene, midges from common buckthorn (*R. cathartica*) and *R. saxatilis* ssp. *tinctorius* are clearly two closely related but distinct species. This further confirms the likely high degree of host specificity of *W. krumbholzi.* CABI was unable to do host-specificity testing since they did not succeed in obtaining reproducing trees of the host, *R. cathartica,* when grown in pots or fruiting trees of other test species. CABI finds it will not be feasible to successfully screen *W. krumbholzi* in the near future. Without host-specificity testing, *W. krumbholzi* could not be approved for release.

A study found no evidence of negative plant–soil feedback by mature *R. cathartica* on conspecifics that could explain low seedling numbers of *R. cathartica* in the native range. There was however a positive plant–soil interaction in the rate of seedling emergence. A small difference in the number of days to seedling emergence probably explains most of the variation in seedling growth.

Due to the difficulties surrounding currently studied agents and the low probability of finding additional potential agents, it has been decided that the project will be stopped and we conclude that there are not suitable biological control insects for *R. cathartica* at this time.

**Result/Activity 2:** Introduction and evaluation of Garlic Mustard biological control agents in MN

**Description:** Activities will include selection of potential release sites, collection of pre-release plant community data, development of rearing methods for control agents, introduction of control agents and initial evaluation of establishment of agents. In anticipation of biological control agents becoming available for garlic mustard, 12 field sites have been selected in different habitat types to implement a biological control program in Minnesota. At these chosen sites, we will continue to collect data on the abundance of both garlic mustard and native plants prior to release, to establish a baseline for assessing the long-term impact of introduced biological control insects. Work will also take place to develop rearing methods for control agents. Once biological control insects are introduced, we will evaluate insect establishment and plant community response to the biological control.

| **Summary Budget Information for Result/Activity 2:** | **ENRTF Budget** | **$150,000** |
|---|---|---|
| | **Amount Spent** | **$150,000** |
| | **Balance** | **$0** |

| Deliverable/Outcome | Completion Date | Budget |
|---|---|---|
| Introduction of first biological control agent | 2/28/11 | $20,000 |
| Monitor release sites; implement rearing | 9/30/11 | $40,000 |
| Insect rearing protocol completed | 2/28/12 | $30,000 |
| Monitor release sites; implement rearing | 9/30/12 | $40,000 |
| Final report with findings and recommendations | 6/30/13 | $20,000 |

**Completion Date: 6/30/14**

**Results Status as of (12/30/13):**
A contract was written with the University of Minnesota for the balance of the funds remaining. Monitoring of garlic mustard plots was conducted in October 2013. Data was entered and submitted.

At the annual meeting of the Technical Advisory Group for Biological Control Agents of Weeds (TAG) group in June, 2013, it was recommended that we include additional Threatened and Endangered (T and E) plants on the Federal list of Threatened and Endangered Species in our test plant list for the potential crown-boring bicontrol insect, *Ceutorhynchus scrobicollis.* There are currently 35 T and E and 7 candidate species in the Brassicaceae family that are listed by the USFWS. To further define the host specificity of *C. scrobicollis*, 7 T and E, one candidate and 6 surrogate species have been identified for further testing. The surrogate species represent T and E species which cannot be tested directly since seed are not available. When we selected surrogates for testing, taxonomically related species were chosen with similar life histories, habitats or ranges as the listed species. With the addition of these Brassicaceae species, we will have tested T and E, candidate species or surrogates from all of the Brassicaceae genera on the USFWS Federal List of Threatened and Endangered Species.

In October, 2013, we submitted a "Proposed Supplemental Test Plant List" based on reviewers' concerns arising from our TAG petition, as well as comments received from the June 2013 TAG meeting. We anticipate a TAG response to our supplemental test plant list in the spring of 2014.

**Results Status as of (6/30/13):**

Garlic mustard field monitoring was conducted in October 2012 and June 2013. Lab experiments were conducted to develop the most efficient and consistently reliable methods to rear *C. scrobicollis* from garlic mustard plants. *Ceutorhynchus scrobicollis* has been successfully reared on caged garlic mustard plants in a growth chamber by alternating growth chamber temperatures and photoperiods to mimic natural conditions in its native range. In Germany, *C. scrobicollis* produces one generation per year and F-1 adults emerge in late May. Simulating a three-month summer aestivation period, followed by a week of fall, and three weeks of winter resulted in optimum levels of oviposition. After receiving shipments of *C. scrobicollis* from Europe, it will be necessary to rear a minimum of one generation in a containment facility to ensure that the endoparasitoid, *Perilitus conseutor*, is not introduced along with adult *C. scrobcollis*. A method was developed to rear parasitoid-free *C. scrobicollis.* A bill is expected from U of M after June 30, 2013. At that point, we will write a contract with the University of MN for the remaining fund amount.

In May and June 2013 we received communication from the USDA APHIS Technical Advisory Group regarding the petition for release of *C. scrobicollis*. The petition was rejected based on the desire to see additional Threatened and Endangered mustard species undergo host-specificity testing. Dr. Katovich and Dr. Becker will work with the US Fish and Wildlife Service on developing a list of species to be tested and obtaining the seeds. At this time, we do not know the final number of additional species that need to be tested, but it appears that it will be fairly small as many of the species have already been tested. The TAG chair indicated that there was strong support for *C. scrobicollis* as garlic mustard biocontrol, but that the additional host-specificity testing was necessary. We will work with TAG and USFWS to obtain an agreed upon list of species to be tested and work with them to obtain seeds.

**Result Status as of (9/30/12)**

There has been no notification from the USDA Technical Advisory Group (TAG) as to the status of the petition for the release of *Ceutorhynchus scrobicollis* as a biological control insect for garlic mustard.

Garlic mustard monitoring data was collected from all 12 sites in June 2012. Data has been entered and research analysis is beginning. Among all 240 plots, the mean garlic mustard percent cover was 19%. Garlic mustard seedling density averaged 74 seedlings/$m^2$ and garlic mustard adult density averaged 12 adults/$m^2$.

Work continues on updating and revising a manual for propagating garlic mustard and rearing *C. scrobicollis*. Progress has been made in establishing rearing methods for the biocontrol insects. In Europe, *C. scrobicollis* adults emerging in the spring require a summer aestivation period before adult females are able to lay eggs. Researchers conducted a study to determine the minimum length of summer aestivation required for adult females to reach maturity and lay eggs when reared in growth chambers in the containment facility. By using the shortest length of aestivation required, insects can be reared more quickly and then more can be produced over time. Results showed that total numbers of eggs per leaf were highest with the standard 3 month aestivation period as opposed to the 1 or 2 month aestivation periods.

Rearing *C. scrobicollis* in the containment facility has not been reliable and researchers have not been able to consistently rear adults on garlic mustard plants. Since *C. scrobicollis* pupate in the soil, it is possible that pupae had low rates of survival in the soil mixes used to propagate garlic

mustard. For this reason, researchers tested whether the addition of 3 to 4 cm of a standard greenhouse soil mix added to the top of a soil-less greenhouse mix would affect the number of *C. scrobicollis* adults emerging from garlic mustard soils. The results of the study showed a significantly greater number of F1 adults emerged from plants with 3 to 4 cm of greenhouse soil mix placed over the soil surface. The conclusion was that the addition of greenhouse soil can aid in *C. scrobicollis* pupal survival. For future *C. scrobicollis* rearing efforts, soil will be added to the top of the peat-based mix prior to placing adults on garlic mustard plants.

**Result Status as of (2/28/12)**
There has been no notification from the USDA Technical Advisory Group (TAG) as to the status of the petition for the release of *Ceutorhynchus scrobicollis* as a biological control insect for garlic mustard.

Garlic mustard monitoring data was collected in October 2011 at all 12 sites. Data was entered and data analysis of the 2011 data has begun. For the 12 sites, the average percent cover of garlic mustard in June 2011 ranged from 6% garlic mustard cover at the lowest cover site to 65% garlic mustard cover at the highest cover site. Average garlic mustard cover at the sites in October 2011 ranged from 0% to 13% cover of garlic mustard. For the 12 sites, the average density of adult stems of garlic mustard ranged from 1-40 stems/$m^2$ in June 2011, the average density of garlic mustard seedlings ranged from 46-655 seedlings/$m^2$ in June 2011, and the average density of garlic mustard rosettes ranged from 0-58 rosettes/$m^2$ in October 2011.

**Result Status as of (9/30/11)**
The petition for release of the garlic mustard biocontrol insect *Ceutorhynchus scrobicollis* was submitted to the USDA Technical Advisory Group (TAG) on Sept. 8, 2011. At this time no insects are approved for garlic mustard biocontrol release. We await the recommendation from TAG. From March-September 2011, work continued to focus on monitoring garlic mustard plots. Data was collected on the 12 permanent garlic mustard monitoring plots in June 2011. 2011 data has been collected and entered, but has not been analyzed.

Results of garlic mustard monitoring in 2010 showed that garlic mustard population density in 2010 was similar to previous years in showing high variability among sites. Garlic mustard is decreasing at two sites which have received management (Luce Line and Pine Bend Bluffs). At Luce Line, herbicide applications have resulted in a decrease in garlic mustard. At Pine Bend Bluffs, cutting trees and converting the site from a forest to savannah has resulted in a decrease in garlic mustard. A common pattern for other garlic mustard sites is for cycling where one life stage (seedling or adult) to dominate in any given year, then the next year, the other life stage dominates. In 2010, three sites showed strong population cycling with the sites alternating between being dominated by the seedling/rosette 1$^{st}$ year life stage in one year and then dominated by the adult 2$^{nd}$ year life stage the next. Three sites showed some cycling, but not consistently. These sites had declines in adult plants in 2009, followed by an increase in 2010. Three sites had increasing garlic mustard from 2005-2008, but now the populations are beginning to cycle and hold steady. One site is showing a decline in garlic mustard.

**Result Status as of (02/28/11):**

A contract was written with the University of Minnesota to carry out the LCCMR funded research on garlic mustard in fall 2010. The main goal was to continue monitoring established permanent plots to monitor garlic mustard populations in anticipation of biological control insect release. From 2005-present, monitoring sites have been surveyed twice yearly with data collected on garlic mustard population density, percent cover, insect damage, and heights and numbers of siliques of the second year plants. In October 2010 data was collected on the garlic mustard monitoring plots. Monitoring data from June and October 2010 is being analyzed and summarized. No biological control agents have been approved for release in the US at this time.

**Final Report Summary:**
Four *Ceutorhynchus* weevil species are being studied to determine their suitability as biological control agents for garlic mustard. Petitions for release have been submitted to the USDA-APHIS Technical Advisory Group (TAG) starting in 2008, but TAG has requested additional host-specificity testing over time. No biological control insects for garlic mustard have been approved for release as of 2014.

In order to develop *C. scrobicollis* as a biocontrol agent for garlic mustard, it was necessary to design reliable and consistent methods to rear the weevils. Studies were conducted to develop mustard propagation methods and *C. scrobicollis* rearing protocols in our High Containment facility the University of Minnesota in anticipation of permission to release *C .scrobicollis* into the field for the biocontrol of garlic mustard. The experiments that were conducted allowed the development of efficient and consistently reliable methods to rear *C. scrobicollis* from garlic mustard plants.

A second focus of research for this report has been monitoring garlic mustard populations in Minnesota to collect pre-release data so efficacy of biocontrol can be measured once insects are released. Long-term monitoring shows that garlic mustard populations can fluctuate widely from year to year. To monitor garlic mustard populations we used a nationally standardized protocol in which data is collected on garlic mustard population density and cover, garlic mustard plant heights and silique (seed pod) production, insect damage to garlic mustard, the cover of the associated plant community, and litter cover. Twenty permanent $0.5m^2$ monitoring plots were established at 12 sites throughout Minnesota. Data was collected each June and October from 2005 to 2013. Nine years of monitoring data show that garlic mustard is currently experiencing very little herbivory in Minnesota and that garlic mustard populations can vary considerably from year to year. As of 2013, garlic mustard is still present in almost all of the plots. Garlic mustard cover is negatively correlated with cover of other species.

## V. TOTAL TRUST FUND PROJECT BUDGET:

**Contract Services: $300,000** (CABI for buckthorn research; and Univ. of MN for garlic mustard implementation)

**TOTAL ENRTF PROJECT BUDGET: $300,000**

## VI. PROJECT STRATEGY:

### A. Project Partners:

Dr. Andre Gassmann, CABI Europe-Switzerland, Delemont, Switzerland will be under contract to continue the ongoing buckthorn research ($150,000). CABI has been working on buckthorn biological control since 2001. CABI is responsible for research on purple loosestrife bio-control agents and many leafy spurge bio-control agents that are currently used in the U. S. and Canada.

Drs. David Ragsdale, Roger Becker and Elizabeth Stamm Katovich, University of Minnesota, will carry out garlic mustard biological control research under contract ($150,000). This amount may change based on future role of Minnesota Department of Agriculture; see below). Drs. Becker and Ragsdale will spend 5% and of their time on this project. Dr. Katovich will spend 60% of her time on garlic mustard.

Monika Chandler, MN Department of Agriculture, will work closely with DNR staff to rear biological control agents and implement evaluations of garlic mustard biological control in the field. Ms. Chandler will spend 5% of her time (in-kind) on this project.

### B. Project Impact and Long-term Strategy:

Development and implementation of biological control for buckthorn could take up to ten years. This research will determine whether there are suitable bio-control agents, whether further research into these potential agents is warranted, and make recommendations for future work. If potential control agents are found, further research would be needed to continue screening the insects to ensure they are host specific and won't feed on other plants. Several insects for garlic mustard control are near completion of host specificity testing and one or more species are expected to be approved for introduction in the United States in 2010. Our time will be spent over the next 5-7 years evaluating the success of the insects introduced. Both European buckthorn and garlic mustard biological control efforts will follow research processes similar to those used for highly successful purple loosestrife and leafy spurge programs that have been funded through the LCCMR process.

### C. Other Funds Proposed to be Spent during the Project Period:

An estimated $3,500 in-kind directly related to this project (e.g. general fund-supported project manager staff time) is expected to be contributed to this project (but not tracked for reporting purposes). Approximately $42,000 in Department Operations and Division Support charges accruing to this project will be covered by Division general funds or other eligible Division funds (see Attachment B.)

Buckthorn related spending: The Department of Natural resources will contribute approximately $30,000 in additional funding towards this project.

### D. Past Spending:

Buckthorn related spending: The DNR spent $20,000 in 2001 to initiate research on buckthorn bio-control. The DNR received $125,000 from the U.S. EPA (2001-2005) to continue the buckthorn research. LCMR funding $109,000 (2003) and $110,000 (2005) recommended funding along with an additional $30,000 from the United States Fish and Wildlife Service (through Minnesota Department of Natural resources) is being used to

continue this research. The Department of Natural Resources contributed an additional $30,000 in 2007.

Garlic mustard related spending: The DNR spent $25,000 in 1999 supporting garlic mustard biological control research. Between 2002 and 2008, the DNR received $265,000 from the U.S.D.A.-Forest Service to continue host specificity testing of garlic mustard agents. LCCMR funded $90,000 (2005) and 135,000 (2007) for garlic mustard research.

**VII. DISSEMINATION**: It is expected that the results of this project will be published in peer-reviewed scientific journals and also in special publications and newsletters. Results also will be presented at national, regional and state scientific meetings to peers in the field, as well as to resource managers and planners who will use the results of this project.

**Buckthorn biological control research has been disseminated in the following ways:**
*Peer reviewed journal publication (pdf attached):*

- Gassmann, A. and I. Tosevski. 2014. Biological control of Rhamnus cathartica: is it feasible? A review of work done in 2002–2012. Journal of Applied Entomology 138: 1-13.

*CABI Report Summary (pdf attached):*

- Gassmann, A., A. Leroux, M. Bennett, M. Penic, N. Haefliger, R. Eschen, J. Jović and I. Toševski. 2012. Report 2010–12: Biological control of common buckthorn, *Rhamnus cathartica*. CABI Europe-Switzerland. CABI Ref: VM01730.

*Poster presentations at conferences:*

- Gassman, Andre, Laura C. Van Riper*, and Luke C. Skinner. Conclusions from 11 Years of Buckthorn Biocontrol Research. Ecological Society of America Conference. 4-9 August 2013. La Crosse, WI.
- Gassman, Andre, Laura C. Van Riper*, and Luke C. Skinner. Conclusions from 11 Years of Buckthorn Biocontrol Research. Upper Midwest Invasive Species Conference. 29-31 Oct 2012. La Crosse, WI.
- Gassman, Andre, Laura Van Riper*, and Luke C. Skinner. Developing Biological Control for Common and Glossy Buckthorn. Invasive Plants Symposium, Dec. 2011. Milwaukee, WI.
- Gassman, Andre, Laura Van Riper*, and Luke C. Skinner. Developing Biological Control for Common and Glossy Buckthorn. Minnesota-Wisconsin Invasive Species Conference, 2-10 Nov 2010. St. Paul, MN.

*Webpage created on MN DNR website:*

- http://www.dnr.state.mn.us/invasives/terrestrialplants/woody/buckthorn/biocontrol.html

**Garlic mustard biological control research has been disseminated in the following ways:**
*Peer reviewed journal publication (pdf attached):*

- Becker, R.L., E.J.S. Katovich, H.L. Hinz, E. Gerber, D.W. Ragsdale, R.C. Venette, D.N. McDougall, R. Reardon, L.C. Van Riper, L.C. Skinner, and D.A. Landis. 2013. The Garlic Mustard (*Alliaria petiolata*) Case, What Makes a Good Biological Control Target. The Intersection of Science, Perspectives, Policy and Regulation. pp. 332-339 *In* Proc. XIII International Symposium on Biological Control of Weeds (ISBCW). Sept. 11-16, 2011. Waikoloa, Hawaii. Wu, Yun; Johnson, Tracy; Sing, Sharlene; Raghu, S.; Wheeler, Greg;

Pratt, Paul; Warner, Keith; Center, Ted; Goolsby, John; and Reardon, Richard, Editors. USDA Forest Service, FHTET-2012-07. January 2013. 536 p. http://www.invasive.org/publications/xiiisymposium/

*U.S. Forest Service Technology Transfer document:*

- Becker, Roger, Esther Gerber, Hariet L. Hinz, Elizabeth Katovich, Brendon Panke, Richard Reardon, Mark Renz, and Laura Van Riper. 2013. Biology and Biological Control of garlic Mustard. US Forest Service Forest Technology Enterprise Team publication FHTET-2012-05. http://www.fs.fed.us/foresthealth/technology/pdfs/GarlicMustardBiocontrol_FHTET-2012-05.pdf [Accessed May 2014].

*Reports to the Environment and Natural Resources Trust Fund (pdfs attached):*

- Katovich, E.J. and Becker, R.L. 2014. Garlic mustard biological control: Developing biological control insects, working towards field release.
- Van Riper, L.C. and Becker, R.L. 2014. Garlic mustard (*Alliaria petiolata*) monitoring in Minnesota: 2005-2013.

*Presentations:*

- E. J. S. Katovich. Upper Midwest Invasive Species Conference. November, 2010. St. Paul, MN. Biocontrol of Garlic Mustard and Buckthorn, an Update.
- E. J. S. Katovich. XIII International Symposium on Biological Control of Weeds. September, 2011. Waikoloa, Hawaii. Biological Control of Garlic Mustard, *Alliaria petiolata*, with the Root and Crown- Boring Weevil, *Ceutorhynchus scrobicollis*.
- E. J. S. Katovich. Ontario Invasive Plant Council, Annual General Meeting and Conference. October, 2011. Picton, Ontario. Potential for the Biological Control of Garlic Mustard.
- E. J. S. Katovich. Upper Midwest Invasive Species Conference. October, 2012. La Crosse, WI. Biological Control of Garlic Mustard with a Seed-Feeding Weevil.
- E. J. S. Katovich. Biological Control of Northeastern Weeds-2013 Cooperators Meeting. February, 2013. Trenton, New Jersey. Garlic Mustard Biocontrol: Current Status and Future Directions.
- E. J. S. Katovich. Technical Advisory Group For the Biological Control of Weeds, Annual Meeting. June, 2013. Washington, D.C. *Ceutorhynchus scrobicollis* as a Potential Biocontrol Agent of Garlic Mustard, *Alliaria Petiolata*.
- E. J. S. Katovich. University of Minnesota, guest lecturer for AGRO 4505: Biology, Ecology and Management of Invasive Plants. Biological Control of Invasive Plants. 2010-2014.

**VIII. REPORTING REQUIREMENTS:** Periodic work program progress reports will be submitted not later than February 2011, September 2011, February 2012, September 2012, June 2013, and December 2013. A final work program report and associated products will be submitted by June 30, 2014.

**Final Attachment A: Budget Detail for 2010 Projects**

**Date: August 15, 2014**

**Project Title: Biological Control of European Buckthorn and Garlic Mustard (111-D)**

**Project Manager Name: Laura Van Riper**

**Trust Fund Appropriation: $ 300,000**

1) See list of non-eligible expenses, do not include any of these items in your budget sheet

2) Remove any budget item lines not applicable

| **2010 Trust Fund Budget** | **Result 1 Budget:** | **Amount Spent** *(08/15/14)* | **Balance** *(08/15/14)* | **Result 2 Budget:** | **Amount Spent** *(08/15/14)* | **Balance** *(08/15/14)* | **TOTAL BUDGET** | **TOTAL BALANCE** |
|---|---|---|---|---|---|---|---|---|
| | Buckthorn biological control - Europe | | | Garlic Mustard biological control | | | | |
| **BUDGET ITEM** | | | | | | | | |
| **Contracts** | $150,000.00 | $150,000.00 | $0.00 | $150,000.00 | 150,000.00 | $0.00 | $300,000.00 | $0.00 |
| **Professional/technical** *(with whom?, for what?)* | CABI Europe-Switzerland: Research in Europe | $130,338.00 | | University of Minnesota: Research in Minnesota | 150,000.00 | | | |
| | University of Minnesota: Research in Minnesota | $19,662.00 | | | | | | |
| **COLUMN TOTAL** | **$150,000.00** | **$150,000.00** | **$0.00** | **$150,000.00** | **$150,000.00** | **$0.00** | **$300,000.00** | **$0.00** |