2005 Project Abstract
For the Period Ending June 30, 2008

PROJECT TITLE: Biological Control of European Buckthorn and Garlic Mustard
PROJECT MANAGER: Luke Skinner
AFFILIATION: Minnesota Department of Natural Resources
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FUNDING SOURCE: Minnesota Environment and Natural Resources Trust Fund
LEGAL CITATION:
[ML 2005, First Special Session, [Chap. 1], Art. 2, Sec.[11], Subd. 5 (h).]

APPROPRIATION AMOUNT: $200,000

Overall Project Outcome and Results
This project builds upon and continues work begun from a 2003 Trust Fund appropriation and has since received an additional 2007 Trust Fund appropriation to further continue and accelerate the work.

Buckthorn and garlic mustard are invasive species of highest priority for development of long-term management solutions, such as biological control (bio-control). This research aimed to help determine 1) if there are suitable insects that can be used to reduce impacts caused by buckthorn and 2) to implement introduction of insects to control garlic mustard and assess their establishment and success.

Buckthorn. Insects were collected and reared for carrying out host specificity testing. A total of 1,733 specimens (356 species) were collected from buckthorn infestations in this insect fauna survey. In total, 39 specialized arthopods were recorded from R. cathartica (common buckthorn) and F. alnus (glossy buckthorn) in Europe.

The reassessment of the potential for biological control of R. cathartica and F. alnus was conducted based on work done in Europe from 2002-2007 on potential biological control agents. A summary of 10 priority species for future research on biological control of R. cathartica is provided in Appendix A of the Work Program Final Report. This final suite of priority species are being tested for use as effective bio-control agents in future work.

Garlic mustard. Pre-release data is providing a greater understanding of normal year-to-year variation. To help differentiate normal fluctuation from changes due to the bio-control insect, data was collected over the course of this project. On average, less than 2% of the leaf area was damaged by herbivores. Garlic mustard plant populations do vary considerably from year to year. Two to three years of pre-release monitoring data have given us a good understanding of the year-to-year fluctuations in populations. At some sites, the population fluctuations are due to the changes in dominance between the seedling and adult stages.

After biological control insects are released we expect to see decreases in garlic mustard populations. With long-term data collection we can see long-term trends in garlic mustard populations (see Appendix B of Work Program Final Report).

Project Results Use and Dissemination

Information garnered from this study will be used to further our objective of developing an effective and efficient bio-control agent for buckthorn and garlic mustard. Effective bio-control agents will help reduce the damage and cost related to control of these invasive species. The information provided by this work helps to establish basic biological information pertaining to the types of species available for potential bio-control agents for buckthorn and narrow our efforts to a few priority species. The information gained on garlic mustard growth and impacts on native species will help us to assess the effectiveness of the current bio-control agents once they have been applied to the test sites. Without this type of baseline data a true understanding of the impacts the bio-control agent is having are impossible to attain. Information from these projects are being shared with multiple federal and state agencies to help the region better understand the potential control mechanisms for buckthorn and garlic mustard.

Information on this work has also been developed into peer reviewed scientific papers. The information has been presented at a variety of national and international conferences. Locally this information has been presented to a variety of interested practitioners and citizens at local conferences and meeting.

LCCMR 2005 Work Program Final Report

Project Completion and Summary Date: June 30, 2008
LCCMR 2005 Work Program Final Report

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

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

Total Biennial LCMR Project Budget:	**LCMR Appropriation:**	**$200,000**
	Minus Amount Spent:	**$200,000**
	Equal Balance:	**$0**

Legal Citation: ML 2005, First Special Session, [Chap. 1], Art. 2, Sec.[11], Subd. 5 (h).

Appropriation Language: 5(h) Biological Control of European Buckthorn and Garlic Mustard. $100,000 the first year and $100,000 the second year are from the trust fund to the commissioner of natural resources to research potential insects for biological control of invasive European buckthorn species for the second biennium and to introduce and evaluate insects for biological control of garlic mustard. This appropriation is available until June 30, 2008, at which time the project must be completed and final products delivered, unless an earlier date is specified in the work program.

II. AND III. FINAL PROJECT SUMMARY:

Buckthorn and garlic mustard are invasive species of highest priority for development of long-term management solutions, such as biological control. This research will help 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.

Buckthorn. Insects were collected and reared for carrying out host specificity testing. A total of 1733 specimens (356 species) were collected from buckthorn infestations in this insect fauna survey. In total, 39 specialized arthopods were recorded from R. cathartica and F. alnus in Europe.

The reassessment of the potential for biological control of R. cathartica and F. alnus was conducted based on work done in Europe from 2002-2007 on potential biological control agents. A summary of 10 priority species for future research on biological control of R. cathartica is given in Table 1, page 13 Appendix B. This final suite of priority species will be tested for use as effective biocontrol agents over the next three years.

Garlic mustard. Pre-lease data is providing a greater understanding of normal year-to-year variation, to help differentiate normal fluctuation from changes due to the biocontrol insect was collected over the course of this project. On average, less than 2% of the leaf area was damaged by herbivores. Garlic mustard plant populations do vary considerably from year to year. Two to three years of pre-release monitoring data have given us a good understanding of the year-to-year fluctuations in populations. At some sites, the population fluctuations are due the changes in dominance between the seedling and adult stages.

After biological control insects are released we expect to see decreases in garlic mustard populations. With long-term data collection we can see long-term trends in garlic mustard populations (see Appendix C).

IV. OUTLINE OF PROJECT RESULTS:

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

Description: Researchers from the Center for Applied Bioscience (CABI) in 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 preliminary information of 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 1:	**LCMR Budget**	**$90,000**
	Balance	**$0**

Completion Date: 6/30/08

Final Report Summary: Over the course of this project researchers with CABI have surveyed, collected and tested a variety of insects for potential biocontrol of *R. cathartica* and *F. alnus.* A total of 1733 specimens (356 species) were collected from buckthorn infestations in this insect fauna survey. In total, 39 specialized arthopods were recorded from *R. cathartica* and *F. alnus* in Europe. Selceted species where tested for ability to oviposition on these plants and their choice

of oviposition plants. These species where also tested for their host specificity preference. These tests help to determine the effectiveness and efficiency of these species as biocontrol agents and any risk associated with other native related shrubs.

Once these surveys and tests were completed CABI researchers reassessed the data collected and prioritized the species for further testing. The reassessment of the potential for biological control of *R. cathartica* and *F. alnus* was conducted based on work done in Europe from 2002-2007 on potential biological control agents. A summary of 10 priority species for future research on biological control of *R. cathartica* is given in Table 1, page 13 Appendix B. This final suite of priority species will be tested for use as effective biocontrol agents over the next three years.

Three of the high priority species identified as priority species included *Philereme vetulata* (Lep., Geometridae), *Trichochermes walkeri* (Hom., Triozidae), and *Wachtiella krumbholzi* (Dipt.; Cecidomyiidae). These three species vary in the type of damage they do to *R. cathartica* ranging from the production of galls to attacking the fruits of the shrubs. One general finding is that there are few if any good biological control candidates for *F. alnus* (glossy buckthorn). All the candidates listed in the Table 1 are insects associated with *R. cathartica* (common buckthorn). The final report for Result 1 can be found in two CABI reports included in Appedix B. The first report describes in detail the research carried out on rearing and testing in 2006, and the second report covers work carried out in 2007 including the a reassement of the research completed to date and includes priorities for future work (appendix B.).

Further funding secured for the FY09/FY10 biennium will help to complete the work on these three potential biocontrol agents for *R. cathartica* control.

Result 2: Survey of insects on buckthorn in Minnesota

Description: Surveys will be continued out to determine what insect species currently utilize buckthorn in Minnesota. Such surveys are needed to determine if any native or non-native insect species are currently found on buckthorn or cause damage to buckthorn. Multiple sites will be surveyed periodically throughout the growing season to capture any insect species associated with buckthorn. Any immature insect collected will be allowed to complete development for identification purposes. A representative sample of each insect species collected will be mounted or preserved, and sent to the appropriate taxonomist for proper identification.

Summary Budget Information for Result 2:	**LCMR Budget**	**$20,000**
	Balance	**$0**

Completion Date: 02/28/07

Final Report Summary: The main effort in 2006 was to complete the sorting, pinning and identification of the large number of insects collected in 2004 and 2005. All insects have now been identified and included in data summaries and analysis. A total of 1733 specimens representing 356 species of insects were collected from buckthorn infestations in this insect fauna survey. Hemiptera was the most abundant order. It was followed by Hymenoptera, which consisted mostly of parasitoids simply using buckthorn as a resting spot or searching for their host. This data was analyzed to look at the relationship of habitat type to insect species richness and how this may affect the introduction of a biological control agent. Data indicates that ample feeding niches are available given that most herbivores collected can be classified as generalists. However, the abundance of parasitoids and predators may hinder establishment of potential

biological control agents. Data analysis are complete and we have attached the final report for result 2 to this document (Appendix B).

Result 3: Introduction and evaluation of Garlic Mustard biological control agents in MN

Description: Research activities will include selection of potential release sites, collection of pre-release plant community data, introduction of control agents and initial evaluation of establishment of agents. In anticipation of biological control agents becoming available for garlic mustard, up to 10 field sites will be select in different habitat types to implement a biological control program in Minnesota. At the chosen sites, we will 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. Once biological control insects are introduced, we will evaluate insect establishment and plant community response to the biological control.

Summary Budget Information for Result 3:	**LCMR Budget**	**$90,000**
	Balance	**$0**

Completion Date: 6/30/08

Final Report Summary:

While evidence of insect feeding was widespread the actual amount of leaf damage was low. Across all sites, seasons and years the average amount of leaf area damaged due to insects was 1.8 ± 0.03%. Leaf damage did not vary widely from site to site. The lowest mean leaf removal was 0.95% at Pine Bend in 2006, while the highest was 4.4% at Fort Snelling. When biological control weevils are released it is expected that insect damage, especially windowpane feeding, will increase.

Garlic mustard's biennial life cycle drives some of the changes in garlic mustard cover and population density from year to year. At some sites, one life stage clearly dominates in each year. For example, a site may be dominated by adult flowering plants in spring 2005 and have few seedlings present. In the fall of 2005 there would be few rosettes. In the spring of 2006, the seedling stage would dominate and the site would have many seedling and very few adults. By fall 2006 there would be many rosettes. This pattern is demonstrated in Figure 2 with photos from Baker Park.

Of the 12 sites, six showed a pattern of one life stage dominating each year (Fig. 1). Over three years of monitoring, the rosette population density cycled from low to high to low in some sites and from high to low to high at others (Fig. 1). It is important to take these population cycles into account when analyzing the impacts of biological control insects. A decrease in adult plants from one year to the next may simply be a result in this natural oscillation in life stage dominance. It will take several years of data to separate out natural population cycles from long-term decreases in population.

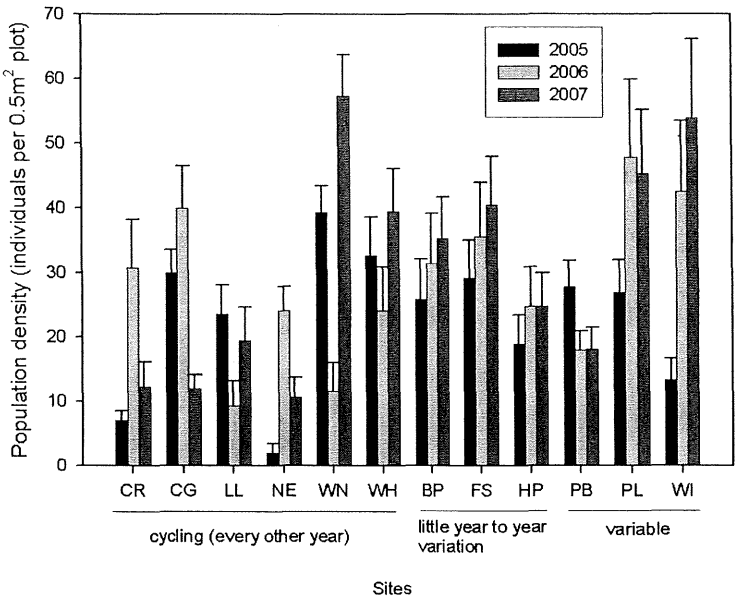

Figure 1. Population density of garlic mustard rosettes over time as measured in the fall at 12 monitoring sites in Minnesota, 2005-2007. Six sites show strong cycling (one life stage is dominant each year) with rosette densities peaking every other year. Three sites show little year to year variation in rosette population density (densities with standard error overlap from year to year). Three sites show variation over time with one site showing a decrease in rosette population density and two sites showing increases in rosette population density.
BP=Baker Park, CR=Coon Rapids, FS=Fort Snelling, HP=Hilloway Park, LL=Luce Line, NE=Nerstrand, PB=Pine Bend, PL=Plainview, WN=Warner Nature, WH=Westwood Hills, WI=Willmar

Data was collected on garlic mustard plant height and number of siliques as measures of vigor and reproductive output of the plants. It is anticipated that the introduction of biological control insects will stress the plants and result in smaller plants which produce fewer siliques. The year to year variation in garlic mustard average heights and numbers of siliques again underscores the importance of pre-release monitoring. Monitoring sites with and without biological control release will help us determine the impacts of biological control agents versus natural year to year variation. Large natural fluctuations in garlic mustard plant height and numbers of siliques were detected as height and siliques production decreased from 2006 to 2007.

One of the impacts of garlic mustard is that it forms dense populations which negatively impact native species. Sites with greater garlic mustard cover had lower native species richness and cover than those sites with less cover of garlic mustard. The negative correlations were consistent in both 2006 and 2007. Sites varied in the amounts of native and nonnative species present. Native species richness ranged from a low of 1.8 species/0.5m^2 quadrat at Baker Park in 2005 to a high of 6.7 species/0.5m^2 at Willmar in 2007. Native species cover ranged from a low of 9% cover at Baker Park in 2005 to a high of 50% cover at Nerstrand in 2007. Nerstrand also had the lowest nonnative species richness and cover (no nonnative species present in the spring 2005-2007). In addition to monitoring whether biological control insects will decrease garlic mustard populations, we can also monitor the response of the native vegetation. Ideally, native species cover and richness will increase as the populations of garlic mustard decrease. Monitoring data provides baseline information on native species cover and richness.

In addition to the proposed monitoring project we also looked at the allelopathic potential of garlic mustard. Garlic mustard roots exude allelochemicals which can negatively affect native

species by decreasing germination rates. The active allelopathic compound in garlic mustard is allyl glucosinolate (sinigrin). The seedbank of the study sites were also examined to provided a better understanding of the restoration potential of the sites infested by garlic mustard. These two portions of the study are discussed in Appendix C.

V. TOTAL LCMR PROJECT BUDGET:

All Results: Other: $200,000 (for contracts)

TOTAL LCMR PROJECT BUDGET: $200,000

VI. OTHER FUNDS & PARTNERS:

A. Project Partners:

Anthony Cortilet and Monika Chandler, MN Department of Agriculture - Mr. Cortilet and Ms. Chandler will work closely with DNR staff to develop and implement evaluations of garlic mustard biological control in the field. Mr. Cortilet will spend ~5% of his time (in-kind) and Ms. Chandler will spend 10% of her time on this project.
Dr. Matthew Cock, Director, Center for Applied Bioscience International (CABI), Delemont, Switzerland - Dr. Cock and his staff will be under contract to continue the ongoing buckthorn research. 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.
Dr. Roger Becker, University of Minnesota, will oversee garlic mustard biological control research under contract. Dr Becker will spend 5% of their time on this project. A post-doctoral researcher will spend 80% of their time on garlic mustard under the direction of Dr. Roger Becker.
Dr. David Ragsdale, University of Minnesota, will carryout surveys for insects on buckthorn in MN. Dr. Ragsdale will spend 5% of his time on this project while his graduate student will spend 50% of their time.
Dr. Bernd Blossey, Cornell University, will provide technical expertise garlic mustard research.

B. Other Funds being Spent during the Project Period:

Buckthorn related spending: The Department of Natural resources will contribute approximately $22,500 in additional funding towards this project.

C. Required Match (if applicable): Not applicable

D. Past Spending:

Buckthorn related spending: The DNR spent $20,000 in 2001 to initiate research on buckthorn bio-control. The DNR received $75000 in 2001 from the U.S. EPA to continue the buckthorn research. Currently, $109,000 of LCMR recommended funding along with an additional $50,000 grant from the U.S. EPA is being used to continue this research. If this research is successful in identifying potential control agents, future proposals will be forthcoming. We will continue to pursue other funding sources for this effort from other

states and federal agencies, which are likely to help pursue bio-control agent if some are identified.

Garlic mustard related spending: The DNR spent $25,000 in 1999 supporting garlic mustard biological control research. Between 2002 and 2003, the DNR received $105,000 from the U.S.D.A.-Forest Service to continue host specificity testing of garlic mustard agents. This research is taking place at the new quarantine facility at the University of MN, St. Paul Campus

E. Time:

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 2006 or 2007. 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 LCMR process.

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.

VIII. REPORTING REQUIREMENTS: Periodic work program progress reports will be submitted not later than January 2006, August 2006, February 2007, August 2007 and March 2008. A final work program report and associated products will be submitted by June 30, 2008.

IX. RESEARCH PROJECTS: See Appendix B and C.

Appendix A: Budget Detail for 2005 Projects

Proposal Title: Biological Control of European Buckthorn and Garlic Mustard-Continuation (H-02)

Project Manager Name: Luke Skinner

LCMR Requested Dollars: $ 200,000

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**

2005 LCMR Proposal Budget	**Result 1, Activity 1 Budget:**	**Amount Spent** 06/30/08	**Balance** 06/30/08	**Result 1 Activity 2 Budget:**	**Amount Spent** 06/30/08	**Balance** 06/30/08	**Result 2 Budget:**	**Amount Spent** 06/30/08	**Balance** 06/30/08	
	Buckthorn biological control- Europe			Buckthorn biological control- MN			Garlic Mustard biological control.			
BUDGET ITEM										TOTAL FOR BUDGET ITEM
Contracts	$ 90,000	$ 90,000	$ -	$ 20,000	$ 20,000	$ -	$ 90,000	$ 90,000	$ -	$ 200,000
Professional/technical	**CABI-Bioscience Switzerand - research in Europe**			**Univ of MN- Research in MN**			**Univ of MN- Research in MN**			
COLUMN TOTAL	$ 90,000	$ 90,000	$ -	$ 20,000	$ 20,000	$ -	$ 90,000	$ 90,000	$ -	$ 200,000