

2007 Project Abstract

For the Period Ending June 30, 2010

PROJECT TITLE: Biological Control of European Buckthorn and Garlic Mustard

PROJECT MANAGER: Luke Skinner

AFFILIATION: Minnesota Department of Natural Resources

MAILING ADDRESS: 500 Lafayette Road, Box 25

CITY/STATE/ZIP: St. Paul, MN 55155-4025

PHONE: 651-259-5140

E-MAIL: luke.skinner@state.mn.us

WEBSITE: [If applicable]

FUNDING SOURCE: Environment and Natural Resources Trust Fund

LEGAL CITATION: M.L. 2007, Chap. 30, Sec.2 Subd. 4(i).

APPROPRIATION AMOUNT: \$300,000

Overall Project Outcome and Results

Garlic mustard (*Alliaria petiolata*) and European/common buckthorn (*Rhamnus cathartica*) are non-native invasive plants that severely threaten native plant communities and degrade wildlife habitat. This project focused on development of biological control as a long-term management strategy for these species. Reports describing the garlic mustard and buckthorn research in detail are attached to this document. Garlic mustard biocontrol agents have not yet been approved for release in the US. Garlic mustard research focused on monitoring the 12 field sites for pre-release research. Garlic mustard monitoring data from 2005 to 2009 showed that garlic mustard populations can vary considerably from year to year. Garlic mustard plants are occurring at high population densities (mean densities up to 133 adult plants/m² and 720 seedlings/m²) and are currently experiencing very little herbivore attack in Minnesota. Work will continue on monitoring the field sites, developing rearing methods, and conducting field releases once insects are available. Buckthorn biocontrol research carried out in 2007–09 concentrated on a leaf-feeding moth, a leaf-margin gall psyllid, and a seed-feeding midge as potential biocontrol agents. The moth was found to lack enough host-specificity and was eliminated from consideration as a biocontrol agent. Host-specificity testing will continue for the leaf gall psyllid as larvae did not develop on the North American *Rhamnus* species tested. One complication is that the phytoplasma '*Candidatus* Phytoplasma rhamni' has been detected in the leaf gall psyllid. Future work will explore the implications of this phytoplasma for using the leaf-gall psyllid as a biocontrol agent. Initial success in rearing a population of the seed-feed midge will allow for future host-specificity testing of this insect. Future work will concentrate on 3 promising potential biocontrol agents, 2 psyllids and the midge.

Project Results Use and Dissemination

The results of the garlic mustard and buckthorn research projects have been shared widely. Updates on the garlic mustard monitoring and biocontrol research and buckthorn biocontrol research were presented at the Minnesota Invasive Species Conference (Oct. 26-29, 2008, Duluth MN). Updates on these projects will be presented at the upcoming Minnesota-Wisconsin Invasive Species Conference (Nov. 8-

10, 2010, St. Paul, MN). In addition, results have been shared across the state through such venues as County Agriculture Inspector meetings, DNR meetings, and Master Gardener meetings. There is considerable interest in these programs and enthusiasm for the potential for biological control of garlic mustard and buckthorn. The results of the garlic mustard monitoring research were reported in the article "Population Biology of garlic mustard (*Alliaria petiolata*) in Minnesota hardwood forests" by L. Van Riper, R. Becker, and L. Skinner in 2010 in the journal *Invasive Plant Science and Management* (3:48-59). Results of the buckthorn research were reported in the article "Use of native range surveys to determine the potential host range of arthropod herbivores for biological control of two related weed species, *Rhamnus cathartica* and *Frangula alnus*" by A. Gassmann, I. Tosevski, and L. Skinner in 2008 in the journal *Biological Control* (45:11-20).

2007 Trust Fund Work Program Final Report

Date of Report: August 16, 2010
Final Report
Date of Work program Approval: June 5, 2007
Project Completion Date: June 30, 2010

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

Project Manager: Luke Skinner
Affiliation: Minnesota Department of Natural Resources
Address: 500 Lafayette Road, Box 25
St. Paul, MN 55155-4025
Telephone number: 651-259-5140
Email: luke.skinner@state.mn.us
Fax: 651-296-1811

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:	\$300,000
	Minus Amount Spent:	\$270,748.38
	Equal Balance:	\$29,251.62

Legal Citation: M.L. 2007, Chap. 30, Sec.2 Subd. 4(i).

Appropriation Language: Biological Control of European Buckthorn and Garlic Mustard. \$300,000 is from the trust fund to the commissioner of natural resources to research potential insects for biological control of invasive European buckthorn species for the third biennium and to introduce and evaluate insects for biological control of garlic mustard for the second biennium. This appropriation is available until June 30, 2010, 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 AND RESULTS:

Garlic mustard (*Alliaria petiolata*) and European/common buckthorn (*Rhamnus cathartica*) are non-native invasive plants that severely threaten native plant communities and degrade wildlife habitat. This project focused on development of biological control as a long-term management strategy for these species. Reports describing the garlic mustard and buckthorn research in detail are attached to this document. Garlic mustard biocontrol agents have not yet been approved for release in the US. Garlic mustard research focused on monitoring the 12 field sites for pre-release research. Garlic mustard monitoring data from 2005 to 2009 showed that garlic mustard

populations can vary considerably from year to year. Garlic mustard plants are occurring at high population densities (mean densities up to 133 adult plants/m² and 720 seedlings/m²) and are currently experiencing very little herbivore attack in Minnesota. Work will continue on monitoring the field sites, developing rearing methods, and conducting field releases once insects are available. Buckthorn biocontrol research carried out in 2007–09 concentrated on a leaf-feeding moth, a leaf-margin gall psyllid, and a seed-feeding midge as potential biocontrol agents. The moth was found to lack enough host-specificity and was eliminated from consideration as a biocontrol agent. Host-specificity testing will continue for the leaf gall psyllid as larvae did not develop on the North American *Rhamnus* species tested. One complication is that the phytoplasma ‘*Candidatus* Phytoplasma rhamni’ has been detected in the leaf gall psyllid. Future work will explore the implications of this phytoplasma for using the leaf-gall psyllid as a biocontrol agent. Initial success in rearing a population of the seed-feed midge will allow for future host-specificity testing of this insect. Future work will concentrate on 3 promising potential biocontrol agents, 2 psyllids and the midge.

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:	Trust Fund Budget	\$165,000
	Amount Spent	\$135,950
	Balance	\$29,050

Completion Date: 6/30/10

Deliverable	Completion Date	Budget
1. Write contract with CABI	2/28/08	\$0
2. Field surveys and collection of agents	8/30/08	\$20,000
3. Host specificity testing completed for 2008	2/28/09	\$40,000
4. Field surveys and collection of agents	8/30/09	\$40,000

5. Host specificity testing completed for 2009	2/28/10	\$45,000
6. Final Report with findings and recommendations	6/30/10	\$20,000

Final Report Summary:

Contracts were written with CABI-Europe Switzerland to carry out the LCCMR funded buckthorn research in Europe. This is in part due to previous contracts with CABI (with funding from the LCMR 2005). This allowed for continuous work on buckthorn biological control without duplication. Additionally, since garlic mustard and the potential biocontrol agents are native to Europe, research did not need to be conducted in a quarantine facility as it would in the United States.

Common buckthorn biocontrol research carried by CABI concentrated on the leaf-feeding moth *Philereme vetulata*, the leaf-margin gall psyllid *Trichochermes walkeri* and the seed-feeding midge *Wachtliella krumbholzi* as potential biocontrol agents. *P. vetulata* was found to lack enough host-specificity and will not be tested further. A few species in the genus *Rhamnus* (*R. alnifolia*, *R. alaternus*, *R. prinoides*) appear suitable hosts for oviposition by *T. walkeri* in no-choice and/or choice conditions but neither gall nor larval development were recorded on any of the North American *Rhamnus* species. The phytoplasma ‘*Candidatus* Phytoplasma rhamni’ has been detected in two populations of *T. walkeri* in Switzerland. *T. walkeri* is the first insect host record for this phytoplasma. Future work will explore the implications of this phytoplasma for *T. walkeri* as a biocontrol agent. Successful oviposition of *W. krumbholzi* was obtained, allowing for future host-specificity testing of this insect. Work will continue on *T. walkeri* and *W. krumbholzi* as they demonstrated enough specificity to warrant further testing.

The full \$165,000 appropriated for this project was not spent. The balance of \$29,050 is the result of a break between contracts with CABI. There was a two month break between contracts as we assembled a panel of 6 outside experts in the fields of biocontrol research, entomology, and pathology. We asked panelists to review the work carried out and provide feedback on future research directions. The panelists were impressed with the work carried out and provided recommendations for future research to focus on *T. walkeri* and the phytoplasma, *W. krumbholzi* host-specificity testing, and causes of buckthorn seedling mortality in Europe (including pathogens).

See the following attached reports for additional information on the buckthorn biocontrol research:

1. “Report 2008-2009: Biological Control of buckthorns, *Rhamnus cathartica* and *Frangula alnus*” by A. Gassmann, I. Tosevski, J. Jovic, N. Guazzone, and D. Nguyen, March 2010.

“Buckthorn Annual Report 2008-09.pdf”

2. Gassmann, A., I. Tosevski, and L. Skinner. 2008. Use of native range surveys to determine the potential host range of arthropod herbivores for biological control of two related weed species, *Rhamnus cathartica* and *Frangula alnus*. Biological Control 45:11-20.

“Gassmann et al 2008 buckthorn.pdf”

Result 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. Unclassified staff will be used by Minnesota Department of Agriculture for the purpose of developing and implementing rearing protocols.

Summary Budget Information for Result 2:	LCMR Budget	\$135,000
	Amount Spent	\$134,798.38
	Balance	\$201.62

Completion Date: 6/30/10

Deliverable	Completion Date	Budget
1. Write contract with University of MN	2/28/08	\$0
2. Monitor garlic mustard field sites	8/30/08	\$10,000
3. Introduction of first bio-control agent	2/28/09	\$30,000
4. Monitor sites; implement rearing	8/30/09	\$40,000
5. Draft of insect rearing protocol completed	2/28/10	\$40,000
6. Final Report with findings and recommendations	6/30/10	\$15,000

Final Report Summary:

Contracts were written with the University of Minnesota to carry out the LCCMR funded research on garlic mustard. The main goal was to continue monitoring established permanent plots to monitor garlic mustard populations in anticipation of biological control insect release. In 2005, 12 garlic mustard sites, spread across eight MN counties, were chosen for long-term monitoring. From 2005-present, 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 addition, data was collected on litter cover and depth and the identity and cover of all other plants in the monitoring plots.

In 2008, a proposal was submitted to USDA-APHIS Technical Advisory Group, petitioning to have the weevil *Ceutorhynchus scrobicollis* approved for release in the United States, to be used as a biological control agent for garlic mustard. In 2009, the Technical Advisory Group recommended additional host-specificity testing of western mustard plant species and a few additional horticultural mustard species. At present, research is continuing to address these concerns and a revised proposal will be submitted in the future. The lack of any biocontrol agents approved for release meant that work had to concentrate on research on garlic mustard populations and not on insect introductions or evaluations of biocontrol establishment or field insect rearing.

Garlic mustard monitoring data from 2005 to 2009 showed that garlic mustard populations can vary considerably from year to year. Garlic mustard plants are occurring at high population densities (mean densities up to 133 adult plants/m² and 720 seedlings/m²) and are currently experiencing very little herbivory in Minnesota. Low herbivory indicates that garlic mustard is not heavily impacted by insects already present in Minnesota and that biocontrol agents could have a large impact. This study examined the fluctuations of garlic mustard

populations over time and their relationship with native species, levels of leaf litter, photosynthetic radiation, and insect herbivores. At half of the 12 monitoring sites, garlic mustard populations showed strong two-point cycling with alternating dominance of the first- and second-year life stages. Increased garlic mustard cover was negatively correlated with native species richness and cover. All sites had litter layers that had been significantly impacted by earthworms. Light was a key factor in understanding garlic mustard populations. Adult plant cover is higher where light is more abundant, but high cover of adult plants produces shade and can cause low cover of seedling plants. We found that less than 2% of garlic mustard leaf area is currently being damaged by herbivores in Minnesota. These results have implications for both the release of potential biological control agents and restoration of garlic mustard invaded sites. When working to restore a site that has been heavily invaded by garlic mustard, the level of earthworm impact, the number and abundance of native species that remain, and any changes to the light available from the canopy should all be considered as factors that could influence the recovery of the site, in addition to the potential decrease in garlic mustard. Future work will focus on monitoring the field sites, developing rearing methods, and conducting field releases once insects are available.

See the following attached reports for additional information on the garlic mustard biocontrol research:

1. “Monitoring garlic mustard (*Alliaria petiolata*) in anticipation of future biocontrol release (2005-2009): Report to the Legislative-Citizen Commission on Minnesota Resources” by L. Van Riper, R. Becker, and L. Skinner, February 2010.
“2010 Garlic mustard monitoring LCCMR report.pdf”
2. . Van Riper, L. C., R. L. Becker, and L. C. Skinner. 2010. Population Biology of garlic mustard (*Alliaria petiolata*) in Minnesota hardwood forests. *Invasive Plant Science and Management* 3:48-59.
“Van Riper et al 2010 garlic mustard.pdf”

V. TOTAL TRUST FUND PROJECT BUDGET:

Contract Services: \$270,748.38 (CABI (buckthorn) and Univ. of MN (garlic mustard))

TOTAL TRUST FUND PROJECT BUDGET: \$300,000

VI. OTHER FUNDS & PARTNERS:

A. Project Partners:

Dr. Andre Gassmann, Center for Applied Bioscience International (CABI), Delemont, Switzerland was under contract to continue the ongoing buckthorn research (\$165,000). 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, Elizabeth Stamm Katovich, and Laura Van Riper, University of Minnesota, carried garlic mustard biological control research under contract

(\$134,904.44). This amount may change based on future role of Minnesota Department of Agriculture; see below). Drs. Becker, Ragsdale, and Katovich spent 10% of their time on this project. Dr. Van Riper spent 100% of her time on garlic mustard.

Monika Chandler, MN Department of Agriculture, was anticipated to work closely with DNR staff to rear biological control agents and implement evaluations of garlic mustard biological control in the field (\$30,000). Ms. Chandler would have spent ~5% of her time (in-kind) on this project. Additional unclassified MDA staff would have been included for implementing rearing of control agents. Funding directed to this activity would have been used once there was approval to remove the control agents from quarantine. Insects were not approved for release during this LCCMR grant period, so MDA staff did not spend time implementing rearing as originally expected.

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

Buckthorn related spending: The Department of Natural resources contributed \$44,871 to pay for indirect costs billed by CABI.

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 \$125,000 from the U.S. EPA (2001-2005) to continue the buckthorn research. Currently, \$110,000 of LCMR 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 will contribute additional funding (up to \$30,000). We will also continue to pursue other funding sources for this effort from other states and federal agencies, which are likely to help pursue bio-control efforts if some potential agents are identified.

Garlic mustard related spending: The DNR spent \$25,000 in 1999 supporting garlic mustard biological control research. Between 2002 and 2006, the DNR received \$225,000 from the U.S.D.A.-Forest Service to continue host specificity testing of garlic mustard agents. LCMR funding (2005) and DNR funding were used as match for a portion of the federal funding. We will continue to use LCCMR recommended funding as match source to leverage Federal funding (up to \$50,000).

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 the next few years. 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.

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.

Results have been published in the following scientific journals:

Van Riper, L. C., R. L. Becker, and L. C. Skinner. 2010. Population Biology of garlic mustard (*Alliaria petiolata*) in Minnesota hardwood forests. *Invasive Plant Science and Management* 3:48-59.

Gassmann, A., I. Tosevski, and L. Skinner. 2008. Use of native range surveys to determine the potential host range of arthropod herbivores for biological control of two related weed species, *Rhamnus cathartica* and *Frangula alnus*. *Biological Control* 45:11-20.

Updates on the garlic mustard monitoring and biocontrol research and buckthorn biocontrol research were presented at the Minnesota Invasive Species Conference (Oct. 26-29, 2008, Duluth MN). Updates on these projects will be presented at the upcoming Minnesota-Wisconsin Invasive Species Conference (Nov. 8-10, 2010, St. Paul, MN). In addition, results have been shared across the state through such venues as meetings of County Agriculture Inspectors, DNR staff, and Master Gardeners.

Supplementary materials submitted in addition to this Work Program Final Report include:

1. Gassmann, A., I. Tosevski, and L. Skinner. 2008. Use of native range surveys to determine the potential host range of arthropod herbivores for biological control of two related weed species, *Rhamnus cathartica* and *Frangula alnus*. *Biological Control* 45:11-20.

“Gassmann et al 2008 buckthorn.pdf”

2. “Report 2008-2009: Biological Control of buckthorns, *Rhamnus cathartica* and *Frangula alnus*” by A. Gassmann, I. Tosevski, J. Jovic, N. Guazzzone, and D. Nguyen, March 2010.

“Buckthorn Annual Report 2008-09.pdf”

3. Van Riper, L. C., R. L. Becker, and L. C. Skinner. 2010. Population Biology of garlic mustard (*Alliaria petiolata*) in Minnesota hardwood forests. *Invasive Plant Science and Management* 3:48-59.

“Van Riper et al 2010 garlic mustard.pdf”

4. “Monitoring garlic mustard (*Alliaria petiolata*) in anticipation of future biocontrol release (2005-2009): Report to the Legislative-Citizen Commission on Minnesota Resources” by L. Van Riper, R. Becker, and L. Skinner, February 2010.

“2010 Garlic mustard monitoring LCCMR report.pdf”

VIII. REPORTING REQUIREMENTS: Periodic work program progress reports will be submitted not later than February 2008, August 2008, February 2009, August 2009 and March 2010. A final work program report and associated products will be submitted by August 16, 2010.

Attachment A: Budget Detail for 2007 Projects - Summary and a Budget page for each partner (if applicable)											
Date: 8/13/2010											
Project Title: Biological Control of European Buckthorn and Garlic Mustard											
Project Manager Name: Luke Skinner											
Trust Fund Appropriation: \$											
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											
2007 Trust Fund Budget	<u>Result 1 Budget:</u>	Amount Spent (date)	Balance (date)	<u>Result 2 Budget:</u>	Amount Spent (date)	Balance (date)	<u>Result 3 Budget:</u>	Amount Spent (date)	Balance (date)	TOTAL BUDGET	TOTAL BALANCE
	Investigate potential insects as biological control of European buckthorn			Introduction and evaluation of Garlic Mustard biocontrol agents in MN			not applicable				
BUDGET ITEM	0		0			0			0	0	0
Contracts			0			0			0	0	0
Professional/technical (with whom?, for what?) Result 1: Contracts with CABI-Europe Switzerland for host-specificity testing of buckthorn biocontrol agents. Result 2: Contracts with the University of Minnesota for garlic mustard research	\$165,000.00	\$135,950.00	\$29,050.00	\$135,000.00	\$134,798.38	\$201.62			\$0.00	\$300,000.00	\$29,251.62
COLUMN TOTAL	\$165,000.00	\$135,950.00	\$29,050.00	\$135,000.00	\$134,798.38	\$201.62	\$0.00	\$0.00	\$0.00	\$300,000.00	\$29,251.62

Use of native range surveys to determine the potential host range of arthropod herbivores for biological control of two related weed species, *Rhamnus cathartica* and *Frangula alnus*

André Gassmann^{a,*}, Ivo Tosevski^a, Luke Skinner^b

^a CABI Europe—Switzerland, Rue des Grillons 1, CH-2800 Delémont, Switzerland

^b Minnesota Department of Natural Resources, 500 Lafayette Road, Box 25, St. Paul, MN 55155-4025, USA

Received 5 June 2007; accepted 19 December 2007

Available online 28 December 2007

Abstract

The buckthorn species, *Rhamnus cathartica* and *Frangula alnus*, are shrubs and small trees of Eurasian origin that have become invasive in North America. A program was initiated in 2001 to reassess the potential for biological control of these two species taking into consideration increasing concerns over potential non-target impacts of biological control agents. The key question was whether *R. cathartica* and *F. alnus* are distantly enough related that they would not share the same arthropod complex in Europe, and, if so, which arthropod species would be less likely to use native North American buckthorns as hosts. Some 1000 insect samples collected at 99 sites in Europe indicated that the arthropod-species richness is higher on *R. cathartica* than on *F. alnus* and includes more species that are presumed to be host-specific at the species or genus level. This discrepancy supports the hypothesis that the genus *Rhamnus* in the temperate Old World has evolved in isolation of the genus *Frangula* in the Neotropics and that taxonomic isolation has an effect on species richness of specialized herbivores. The fauna was dominated by Lepidoptera (22 species), followed by Hemiptera (8 species), Diptera (4 species), Acarina (4 species) and Coleoptera (1 species). At least 12 arthropod species were found exclusively on *Rhamnus*, some of which may be specific to *R. cathartica*. Several species usually associated with *Rhamnus* were found rarely on *F. alnus* but the field host range of these species still needs to be confirmed. Only one species was found exclusively on *F. alnus*. The findings indicate that, with one exception, there are no species or genus-specific agents available for biological control of *F. alnus* at this stage. However, additional field surveys may reveal other host-specific species.

© 2008 Elsevier Inc. All rights reserved.

Keywords: *Rhamnus cathartica*; *Frangula alnus*; Buckthorn; Biological control; Species richness; Food niche; Center of origin; Taxonomic isolation; Host plant phylogeny

1. Introduction

Rhamnus cathartica L. (common buckthorn) and *Frangula alnus* Miller (glossy buckthorn) (Rhamnaceae) are two shrubs and small trees of Eurasian origin which have become invasive in North America.

Rhamnus cathartica is found throughout Europe, but is absent from most parts of Scandinavia and the Iberian Pen-

insula, and from the extreme south (Tutin, 1968; <http://linnaeus.nrm.se/flora/di/ramna/ramn/ramcat.html>). The species is also present in European Russia, in south-western Siberia, in the northern Caucasus as well as in the Province of Xinjiang in China (D. Jianqing, personal communication, 2001). In Europe, *R. cathartica* prefers mesic to mesic-dry, warm open or half-shaded habitats. It grows in well drained calcareous alkaline or neutral soils, but it can also be found occasionally in swampy areas (Rameau et al., 1989).

Rhamnus cathartica was introduced to North America as an ornamental shrub in the late 1800s and was originally used for hedges, farm shelter belts, and wildlife habitats

* Corresponding author. Fax: +41 32 421 4871.

E-mail address: a.gassmann@cabi.org (A. Gassmann).

(Gourley, 1985; Randall and Marnelli, 1996; Gale, 2001). It has spread extensively and is currently found in most Canadian provinces (Nova Scotia to Saskatchewan) and 27 states predominantly in the north central and northeastern portion of the United States (Gale, 2001; USDA/NRCS, 2001). *Rhamnus cathartica* invades mainly woodlands and savannas, although it also occurs on prairies and open fields.

Frangula alnus has a slightly wider distribution than *R. cathartica* extending from northern Scandinavia in the boreal zone up to the Iberian Peninsula and a southernmost enclave in western North Africa (Tutin, 1968; Scamoni, 1985; Medan, 1994; <http://linnaeus.nrm.se/flora/di/rhamna/frang/franaln.html>). *Frangula alnus* is also present in European Russia, in south-western Siberia, in the northern Caucasus as well as in the Province of Xinjiang in China (D. Jianqing, personal communication, 2001). In Europe, *F. alnus* prefers mesic to mesic-moist acid soils in open or half-shaded habitats but it can also be found occasionally in dry calcareous stands (Rameau et al., 1989).

Frangula alnus was imported to North America prior to the 1900s as horticultural stock for landscape plantings, and has become naturalized in the northeastern US and southeastern Canada (Catling and Porebski, 1994; Randall and Marnelli, 1996; Haber, 1997). Currently, *F. alnus* occurs from Nova Scotia to Manitoba, south to Minnesota, Illinois, New Jersey and Tennessee incorporating 23 states in the US (Converse, 2001; USDA/NRCS, 2001). *Frangula alnus* is most problematic in fens and other wetlands but also can invade uplands and sandy soil forests.

Both species are very adaptable, forming dense thickets that shade and inhibit the growth of native forbs, shrubs, and tree seedlings (Heidorn, 1991; Randall and Marnelli, 1996). Both species are alternate hosts for the fungus, *Puccinia coronata* Corda, which causes oat rust disease (Harder and Chong, 1983; Leonard, 2003). *Rhamnus cathartica* and *F. alnus* are also overwintering hosts for the Asian soybean aphid, *Aphis glycines* Matsumura, a pest of soybean, *Glycine max* (L.) Merrill, which was first recorded in North America in 2000 (Voegtlin et al., 2005). American robins (*Turdus migratorius* L.) nesting in *R. cathartica* experience higher rates of predation than conspecifics nesting in native shrubs (Schmidt and Whelan, 1999).

The systematics of buckthorns has a long history of complexity and uncertainty. Linnaeus described *Rhamnus cathartica* and *Rhamnus frangula* in 1753 (Linnaeus, 1753). In 1754, Miller described the genus *Frangula*, and in 1768, transferred glossy buckthorn to this genus under the name *Frangula alnus* (Miller, 1754/1768). The generic recognition of *Frangula* has been disputed and for many years *F. alnus* has gone under the name *R. frangula* L. A recent molecular study by Bolmgren and Oxelman (2004) supports the generic recognition of *Frangula*. The global sample of *Frangula* used in their study represents a well-supported monophyletic sister clade to the rest of *Rhamnus* in its widest sense. Given the lack of resolution in the *Frangula* clade, *R. cathartica* may be considered more distantly

related to the American *Rhamnus* native community than *F. alnus* is to the American native *Frangula* community.

It is difficult to obtain an accurate count of *Rhamnus* and *Frangula* species, in particular in the Old World tropics and eastern Palearctic. Grubov (1949) suggested that *Rhamnus* s.l. consists of almost 200 species but this number was reduced to 125 by Johnston and Johnston (1978). Work by the same authors on Neotropical *Rhamnus* s.l. suggests that *Frangula* has an area of diversification in the mountainous areas of the Neotropics with its southern limit in Northern Argentina. Of the 21 species recognized by Johnston and Johnston (1978) in the Neotropics, 20 belong to *Frangula* and one, *Rhamnus serrata* Humb. and Bonpl. ex J.A. Schultes, to the genus *Rhamnus*. In Europe, the genus *Rhamnus* includes 23 taxa and the genus *Frangula* includes only four taxa (Tutin, 1968; Hampe et al., 2003). In the United States, *Frangula* and *Rhamnus* include five and seven native taxa, respectively, but another two *Rhamnus* subspecies and 10 *Frangula* subspecies have been recorded (USDA/NRCS, 2001). Thus, it appears that *Rhamnus* and *Frangula* are predominant in the Old World and New World, respectively. In North America, the geographical distribution of the two invasive buckthorn species overlaps most with the native species *Rhamnus alnifolia* L'Hér., *Rhamnus lanceolata* Pursh and *Frangula caroliniana* (Walt.) Gray, making these key-species in host range studies of potential biological control agents.

Research to develop biological control for buckthorns was initiated in 1964. Surveys for potential arthropod biological control agents were carried out mostly in Eastern Austria in summer 1964 and 1965 and preliminary screening tests in 1966–1967 (Malicky et al., 1970). A new program was initiated in 2001 to reassess the potential of biological control of buckthorns with regard to the work carried out by Malicky et al. (1970). In recent years there have been ever-increasing concerns over potential non-target impacts of biological control agents and greater demands for high levels of specificity (e.g. Louda et al., 1997; Pemberton, 2000). The key question was whether *R. cathartica* and *F. alnus* are sufficiently distantly related that they would not share the same arthropod complex in Europe and, consequently which arthropod species could be selected for further host range studies, and possibly later on be used for biological control without damaging native North American buckthorns.

The aim of this study was to use both a literature review and field surveys to identify the specialized herbivorous arthropods on *R. cathartica* and *F. alnus* and to determine their host use patterns and preferences in the field. We report results of surveys carried out in Europe in 2002–2005 and review previous studies on the biological control of buckthorns for North America.

2. Materials and methods

Between 2002 and 2005, extensive surveys for presumed-specialized insect species (defined here as a species

restricted to one plant genus) on *Rhamnus* and *Frangula* spp. were carried out in Switzerland, Germany, Italy, the Czech Republic, Austria, Serbia and Montenegro. More intensive surveys were concentrated in the areas which had been sampled in previous years by Malicky et al. (1970), i.e. eastern Austria, Germany and Switzerland. In Serbia and Montenegro, selective surveys were carried out for a few specific insect species that had not been found in the other surveys. With the exception of the Czech Republic and Italy, most sites were sampled twice per season or more, or at different periods of the year for more than 1 year. Fruits were sampled in 2004–05 only. Leaves, stems and fruits were carefully examined for herbivory and symptoms of herbivory. Immature and mature phytophagous arthropods were handpicked or aspirated from young and mature buckthorn plants. In total, we surveyed *R. cathartica* and *F. alnus* at a total of 99 sites and 1000 samples were examined separately. In addition, *Rhamnus alpina* L. and *Rhamnus saxatilis* Jacq. were opportunistically surveyed at two and four sites, respectively, as they can co-occur with *R. cathartica* in the surveyed areas. Juvenile insects were reared on their field host plants in ventilated plastic containers in a shade house. Unhealthy fruits, shoot tips or small branches were collected for dissection or emergence of specific insects. Pheromone traps using a commercial lure developed for *Synanthedon myopaeformis* (Borkh.) by Plant Research International, Wageningen UR, The Netherlands, were used to detect the presence of the root-boring moth *Synanthedon stomoxiformis* Hb. The frequency of occurrence of each arthropod collected was calculated for each buckthorn species surveyed. The sampling unit was the whole habitat/site.

3. Results

In total, 39 specialized arthropods were recorded from *R. cathartica* and *F. alnus* in Europe (Table 1). Lepidoptera (22 species) largely dominated, followed by Hemiptera (8 species), Diptera (4 species), and Acarina (4 species). There was only one specialized beetle species, *Oberea pedemontana* Chevrolat, recorded on these two buckthorn species in Europe. The feeding guild on *R. cathartica* and *F. alnus* was dominated by leaf feeders (18 species), followed by sap-suckers (9 species) and flower or fruit feeders (6 species) of which four species were gall midges reported to induce galls either in the flowers or fruits of buckthorn. In addition to the gall midges, the larvae of *Hysterosia sodaliana* Haw. developed within the fruits of buckthorn and those of *Sorhagenia rhamniella* Zeller lived gregariously between spun blossoms of *R. cathartica*. There were only three shoot/root borers. The larvae of *Sorhagenia janiszewskae* Riedl developed in the shoot-tips and those of *O. pedemontana* in the branches. The larvae of *S. stomoxiformis* mined the roots of buckthorn. Of the leaf feeding phytophages, five species (i.e. *Bucculatrix frangutella* Goetze, *Bucculatrix rhamniella* Zeller, *Calybites quadrisignella* Zeller, *Stigmella catharticella* Stainton and *Stigmella rhamnella*

H.-S.) mined in the leaves of buckthorn partially or during their entire life cycle. Finally, there were three leaf gall forming species, *Trichohermes walkeri* Foerster, *Trioza rhamni* Schrank and *Phyllocoptes annulatus* (Nal.). Comparatively, Brändle and Brandl (2001) found 29 specialists for a total of 91 herbivores on *Frangula* and *Rhamnus* in a study on herbivore species richness on 25 native trees in Germany.

The number of specialized arthropods was much larger on *R. cathartica* than on *F. alnus*. Twenty-two species were mostly associated with *R. cathartica* and other species in the genus *Rhamnus* (Tables 1 and 2). Of these 22 species, only eight species have also been occasionally recorded on *F. alnus* and among those, five consist of literature records, i.e. *S. rhamniella*, *Triphosa sabaudia* Dup., *C. quadrisignella*, *S. rhamnella* and *H. sodaliana*, which were not confirmed during surveys for biological control. However, host affiliations can vary geographically (Fox and Morrow, 1981). For example, *S. rhamniella* is known from *F. alnus* in England (Emmet, 1969), an area that was not surveyed for biological control agents. Another eleven species have been found occurring on both *R. cathartica* and *F. alnus*. Of those, three species, *Ancylis apicella* Den. and Schiff., *Gonopteryx rhamni* L. and *S. janiszewskae* were found more often on *F. alnus* than on *R. cathartica* (Table 2).

The leaf-hopper *Zygina suavis* Rey was the only species found on *F. alnus* but not on *R. cathartica*, although the literature record list *R. cathartica* as a host of *Z. suavis* (Ossiannilsson, 1981). In addition to *Z. suavis*, literature records indicated another five arthropod species known from *F. alnus*, i.e. *Contarinia rhamni* Ruebs., *Dasyneura frangulae* Ruebs., *Lygocoris rhamnicolla* Reuter, *Aristotelia pancaliella* Stgr. and *Eriophyes rhamni* (Pgst.).

Records of presence or absence of most arthropods associated with *R. cathartica* and *F. alnus* in the areas surveyed in Europe in 2002–2005 matched well with those from Malicky et al. (1970) (Table 2). In contrast, the frequency of occurrence of several species differed considerably reflecting a non-random sampling method focussing on pre-selected specialized species in our surveys. *Trichohermes walkeri*, *T. rhamni* and to a slightly lesser extent *Philereme vetulata* Den. and Schiff. and *Triphosa dubitata* L., best represented the specific arthropod community associated with *R. cathartica* in Europe.

Frangula alnus was best represented by *Z. suavis*, *G. rhamni* and *A. apicella* while *B. frangutella* and *O. pedemontana* were recorded equally on both buckthorn species. *Sorhagenia janiszewskae* has been recorded on *F. alnus* and *R. cathartica* in Austria only. The occurrence of this species has not been confirmed on *R. cathartica* in Switzerland and Germany. The frequency of occurrence of most of the other species collected was too low to draw conclusions about host specificity, but our observation matched those of Malicky et al. (1970), indicating that most of the additional species are not associated with *F. alnus* in the areas surveyed.

Table 1

Specialized arthropods associated with *Rhamnus cathartica* and *Frangula alnus* in Europe (*field records from our surveys; **Malicky et al., 1970; remaining records from literature as indicated)

Species	Host plants	Specificity ^a	Food niche	References
Coleoptera				
<i>Cerambycidae</i>				
<i>Oberea pedemontana</i> Chevrolat	<i>R. cathartica</i> *, <i>F. alnus</i> *, <i>R. alpina</i> , <i>Lonicera</i> ?	O?	Stem, woodboring	Horion (1974); Lekic and Mihajlovic (1976); Contarini and Garagnani (1980); Baronio et al. (1988); Demelt and Franz (1990); Frisch (1992)
Diptera				
<i>Cecidomyiidae</i>				
<i>Contarinia rhamni</i> Ruebs.	<i>F. alnus</i>	M	Gall forming (flowers)	Houard (1909); Barnes (1951); Buhr (1965); Zerova et al. (1991)
<i>Dasyneura frangulae</i> Ruebs.	<i>F. alnus</i>	M	Gall forming (flowers)	Barnes (1951); Buhr (1965)
<i>Lasioptera kozarzewskella</i> Mar.	<i>R. cathartica</i>	M	Gall forming (fruits)	Stelter (1975); Zerova et al. (1991)
<i>Wachtliella krumbholzi</i> Stelter	<i>R. cathartica</i> *	M	Gall forming (fruits)	Stelter (1975)
Heteroptera				
<i>Miridae</i>				
<i>Heterocordylus erythrophthalmus</i> Hb	<i>R. cathartica</i> ** , <i>F. alnus</i> **	O	Sap sucking	Gollner-Scheiding (1972)
<i>Lygocoris rhamnicola</i> Reuter	<i>F. alnus</i>	M	Sap sucking	Coulianos (1998)
Homoptera				
<i>Aphididae</i>				
<i>Aphis commensalis</i> Stroyan	<i>R. cathartica</i>	M	Gall forming? (leaves)	Buhr (1965); Heie (1986)
<i>Aphis mammulata</i> Gimingham. & HRL	<i>R. cathartica</i>	M	Sap sucking, free living	Heie (1986); Blackman and Eastop (1994)
<i>Cicadellidae</i>				
<i>Zygina suavis</i> Rey	<i>F. alnus</i> */ <i>R. cathartica</i>	O	Sap sucking, free living	Ossiannilsson (1981)
<i>Psyllidae</i>				
<i>Cacopsylla rhamnicola</i> (Scott)	<i>R. cathartica</i> */**/ <i>F. alnus</i> **	O	Sap sucking, free living	Ossiannilsson (1992)
<i>Triozidae</i>				
<i>Trichochermes walkeri</i> Foerster	<i>R. cathartica</i> */**	M	Gall forming (leaves)	Buhr (1965); Okopnyi and Poddubnyi (1983); Meyer (1987); Zerova et al. (1991); Ossiannilsson (1992); McLean (1993)
<i>Triozia rhamni</i> Schrank	<i>R. cathartica</i> */**/ <i>F. alnus</i> **	O	Gall forming (leaves)	Buhr (1965); Ossiannilsson (1992)
Lepidoptera				
<i>Bucculatricidae</i>				
<i>Bucculatrix frangutella</i> Goeze	<i>R. cathartica</i> */**/ <i>F. alnus</i> */**/ <i>R. alpina</i> */**	O	Leaf miner/leaf chewer	Hering (1957); Heath and Emmet (1985)
<i>Bucculatrix rhamniella</i> H.-S.	<i>R. cathartica</i>	M	Leaf miner/leaf chewer	Hering (1957); Buszko (1992)
<i>Cosmopterigidae</i>				
<i>Sorhagenia lophyrella</i> Douglas	<i>R. cathartica</i> **/ <i>R. saxatilis</i> **	O	Leaf roller	Baran (1997); Malicky et al. (1970)
<i>Sorhagenia janiszewskae</i> Riedl	<i>R. cathartica</i> */**/ <i>R. alpina</i> **/ <i>F. alnus</i> */**/	O	Shoot miner	Malicky et al. (1970)
<i>Sorhagenia rhamniella</i> Zeller	<i>R. cathartica</i> **/ <i>F. alnus</i>	M?	Flower feeder	Malicky et al. (1970); Emmet (1969)
<i>Gelechiidae</i>				
<i>Aristotelia pancaliella</i> Stgr.	<i>F. alnus</i>	M	Leaf chewer	Ivinskis et al. (1982)
<i>Geometridae</i>				
<i>Odontognophos dumetata</i> Treitschke	<i>R. cathartica</i>	M	Leaf chewer	Forster and Wohlfahrt (1981)
<i>Philereme transversata</i> Hufnagel	<i>R. cathartica</i> */**/ <i>R. saxatilis</i> **/ <i>R. orbiculata</i> **/ <i>F. alnus</i> **	O		(Skinner, 1984)
<i>Philereme vetulata</i> Den. and Schiff.	<i>R. cathartica</i> */**/ <i>R. alpina</i> **	O	Leaf chewer	Forster and Wohlfahrt (1981); Skinner (1984)

Table 1 (continued)

Species	Host plants	Specificity ^a	Food niche	References
<i>Triphosa dubitata</i> L.	<i>R. cathartica</i> */**/ <i>R. alpina</i> */**/ <i>F. alnus</i> */**/ <i>Prunus</i> ?/ <i>Fraxinus</i> ?/ <i>Crataegus</i> ?	O?	Leaf chewer	Blaschke (1914); Forster and Wohlfahrt (1981); Skinner (1984); Jacobi and Menne (1991)
<i>Triphosa sabaudia</i> Dup.	<i>R. cathartica</i> */ <i>R. saxatilis</i> */ <i>R. orbiculata</i> */ <i>F. alnus</i> / <i>R. alpina</i>	O	Leaf chewer	Blaschke (1914); Forster and Wohlfahrt (1981)
<i>Gracillariidae</i>				
<i>Calybites quadrisignella</i> Zeller	<i>R. cathartica</i> */**/ <i>F. alnus</i>	M?	Leaf miner/leaf chewer	Hering (1957)
<i>Nepticulidae</i>				
<i>Stigmella catharticella</i> Stainton	<i>R. cathartica</i> */**/ <i>R. alaternus</i>	M?	Leaf miner	Hering (1957); Heath (1976); Speight and Cogan (1979); Puplyasis (1984); Puplesis (1994); Michalska (1996)
<i>Stigmella rhamnella</i> H.-S.	<i>R. cathartica</i> */**/ <i>R. alpina</i> */ <i>F. alnus</i>	O	Leaf miner	Hering (1957); Puplesis (1994); Michalska (1996)
<i>Pieridae</i>				
<i>Gonopteryx rhamni</i> L.	<i>R. cathartica</i> */**/ <i>R. orbiculata</i> */ <i>F. alnus</i> */**	O	Leaf chewer	Frohawke (1940); Bergmann (1952); Pollard and Hall (1980); de Freina (1983); Bibby (1983); Rippey (1984); Heath and Emmet (1989); McKay (1991); Gutierrez and Thomas (2000)
<i>Pyralidae</i>				
<i>Acrobasis romanella</i> Mill.	<i>R. cathartica</i> */ <i>R. alaternus</i> **	O	Lchewer	Malicky et al. (1970)
<i>Trachycera legatea</i> Haw.	<i>R. cathartica</i> */**/ <i>R. saxatilis</i> **	O	Leaf chewer	Mihajlovic (1978)
<i>Sesiidae</i>				
<i>Synanthedon stomoxiformis</i> Hb.	<i>R. cathartica</i> */ <i>F. alnus</i> */ <i>Sorbus aria</i> ?/ <i>Coryllus avelana</i> ?	O?	Root miner	Doczkal and Rennwald (1992); Stadie (1995); Bittermann (1997); de Freina (1997); Spatenka et al. (1999)
<i>Tortricidae</i>				
<i>Ancylis apicella</i> Den. & Schiff.	<i>R. cathartica</i> */**/ <i>F. alnus</i> */**/ <i>R. alpina</i> */**/ <i>Ligustrum</i> ?/ <i>Cornus</i> ?/ <i>Prunus</i> ?	O?	Leaf chewer	Razowski (2003)
<i>Ancylis derasana</i> Hb. (=unculana Haw.)	<i>R. cathartica</i> */**/ <i>F. alnus</i> */ <i>Corylus</i> ?/ <i>Rubus</i> ?/ <i>Populus</i> ?	O?	Leaf chewer	Razowski (2003)
<i>Ancylis obtusana</i> Haw.	<i>R. cathartica</i> */ <i>F. alnus</i>	O	Leaf chewer	Razowski (2003)
<i>Hysterosia sodaliana</i> Haw.	<i>R. cathartica</i> */**/ <i>F. alnus</i>	O	Fruit feeder	Hannemann (1964); Razowski (1970)
<i>Acari</i>				
<i>Eriophyidae</i>				
<i>Aceria rhamni</i> Roiv.	<i>R. cathartica</i>	M	Sap sucker, free living	Amrine and Stasny (1994)
<i>Eriophyes rhamni</i> (Pgst)	<i>F. alnus</i>	M	Leaf erineum ?	Amrine and Stasny (1994)
<i>Phyllocoptes annulatus</i> (Nal.)	<i>R. cathartica</i> *	M	Leaf erineum	Amrine and Stasny (1994)
<i>Tetra rhamni</i> Roiv.	<i>R. cathartica</i>	M	Sap sucker, free living	Amrine and Stasny (1994); Petanovic, personal communication (2005)

^a M, monophagous, restricted to *R. cathartica* or *F. alnus*; O, oligophagous, restricted to species in the genus *Rhamnus* and/or *Frangula*.

In Serbia and Montenegro, *O. pedemontana* and *S. stomoxiformis* were recorded on *R. cathartica* and *F. alnus*, and *Wachtliella krumbholzi* Stelter reared from the fruits of the former species (Gassmann et al., 2006). According to M. Skuhrava (personal communication, 2005), *W. krumbholzi*, which was known from Northern Germany and the Czech Republic only, can-

not be considered to be cecidogenous, but it is rather a seed feeder.

Rhamnus cathartica and *F. alnus* were observed growing sympatrically in 20 sites. Host plant records for 18 arthropod taxa associated with *R. cathartica* and *F. alnus* were similar in allopatric and sympatric sites (Table 3). None of the species known exclusively from *R. cathartica* in allo-

Table 2

Frequency of occurrence of specialized buckthorn arthropods in 2002–05 in Italy, Austria, Switzerland, Germany and the Czech Rep. (records from Malicky et al. (1970) are presented on shaded columns)

	<i>Rhamnus cathartica</i>		<i>Frangula alnus</i>	
Total # of sites sampled :	52	214	47	83
Number of sites with (%):				
LEPIDOPTERA :				
Bucculatricidae :				
<i>Bucculatrix frangutella</i> ⁴⁾	7 (13.5)	44 (20.6)	6 (12.8)	11 (13.3)
Cosmopterigidae :				
<i>Sorhagenia janiszewskae</i> ⁴⁾	13 (25.0)	9 (4.2)	14 (29.8)	14 (16.9)
<i>Sorhagenia rhamnella</i>	-	14 (6.5)	-	-
<i>Sorhagenia lophyrella</i> ³⁾	-	27 (12.6)	-	-
Gracillariidae :				
<i>Calybites quadrisignella</i>	3 (5.8)	10 (4.7)	-	-
Nepticulidae:				
<i>Stigmella catharticella</i>	2 (3.8)	25 (11.7)	-	-
<i>Stigmella rhamnella</i> ⁴⁾	1 (1.9)	2 (0.9)	-	-
Pyralidae :				
<i>Acrobasis romanella</i> ¹⁾	1 (1.9)	--	-	-
<i>Trachycera legatea</i> ³⁾	-	3 (1.4)	-	-
Pieridae :				
<i>Gonopteryx rhamni</i> ^{2) 4) 5)}	7 (13.5)	18 (8.4)	21 (44.7)	22 (26.5)
Geometridae :				
<i>Philereme vetulata</i> ⁴⁾	14 (26.9)	68 (31.8)	-	-
<i>Philereme transversata</i> ^{2) 3)}	8 (15.4)	38 (17.8)	-	1 (1.2)
<i>Triphosa dubitata</i> ^{2) 4)}	22 (42.3)	42 (19.6)	-	3 (3.6)
<i>Triphosa sabaudia</i> ³⁾	-	2 (0.9)	-	-
Tortricidae :				
<i>Ancylis apicella</i> ^{1) 3) 4)}	5 (9.6)	2 (0.9)	7 (14.9)	2 (2.4)
<i>Ancylis derasana</i>	6 (11.5)	9 (4.2)	3 (6.4)	-
Cochylidae:				
<i>Hysterosia sodaliana</i>	-	14 (6.5)	-	-
HOMOPTERA				
Psyllidae :				
<i>Cacopsylla rhamnicola</i>	5 (9.6)	6 (2.8)	-	1 (1.2)
Triozidae :				
<i>Trichoermes walkeri</i>	43 (82.7)	67 (16.8)	-	-
<i>Trioza rhamni</i> ⁴⁾	36 (69.2)	36 (16.8)	-	1 (1.2)
Cicadellidae :				
<i>Zygina suavis</i>	-	-5	(10.6)	-
HETEROPTERA				
Miridae:				
<i>Heterocordylus erythrophthalmus</i>	-	6 (2.8)	-	1 (1.2)
COLEOPTERA				
Cerambycidae:				
<i>Oberea pedemontana</i>	2 (3.8)	-2	(4.3)	-
ACARI				
Eriophyidae				
<i>Phyllocoptes annulatus</i>	9 (17.3)	--		-

¹⁾ Also recorded on *Rhamnus alaternus* (Malicky et al. 1970)

²⁾ Also recorded on *R. orbiculata* (Malicky et al. 1970)

³⁾ Also recorded on *R. saxatilis* (Malicky et al. 1970)

⁴⁾ Also recorded on *R. alpina* (Malicky et al. 1970; personal observations)

⁵⁾ Also recorded on *Frangula rupestris* (Malicky et al. 1970)

patric sites have been recorded on *F. alnus* in sympatric sites, i.e. when given a proximate choice. In sympatric sites, *S. janiszewskae* and *G. rhamni* were the only species that were recorded more often on *F. alnus* than on *R. cathartica*.

Preliminary screening tests with *P. vetulata*, *T. dubitata* and *T. walkeri*, confirmed host plant use observed in the field and the unsuitability of *F. alnus* for insect species associated with *Rhamnus* in their native range (unpublished data). In contrast, both *Rhamnus* and *Frangula* species were suitable hosts for the larvae of *A. apicella* and *S. stomoxiformis*, two species which were recorded on both buckthorns in Europe.

4. Discussion

Assessing the risk to non-target species by a biological control agent has been a fundamental part of classical biological weed control for many decades. When developing biological control for *R. cathartica* and *F. alnus*, minimizing the risk of potential non-target effects might require the selection of agents which are specific to either *R. cathartica* or *F. alnus*. Several of the European arthropod species commonly found in areas that were surveyed during this study are considered monophagous on *R. cathartica*, or oligophagous on species in the genus *Rhamnus*.

Among the leaf chewing species associated with the genus *Rhamnus*, a geometrid, *P. vetulata*, appears to be the most specialised. Among the gall formers and sap-suckers that have been studied so far, the leaf margin gall psyllid *T. walkeri* seems to be monospecific on *R. cathartica*. There is considerable interest in this species because it

attacks *R. cathartica* later in the season than *P. vetulata*. In North America, the high rate of seed production is an important element contributing to the invasiveness of *R. cathartica* (Knight et al., 2007). The seed-feeding midge, *W. krumbholzi* is considered a key candidate biocontrol agent because it could significantly reduce the seed production of common buckthorn in North America. The psyllids *C. rhamnicolla* and *T. rhamni*, the leaf miners *S. catharticala*, *C. quadrisignella* and *B. rhamniella*, the leaf chewers *T. dubitata* and *P. transversata*, and the mites *A. rhamni* and *T. rhamni* are probably specific to *R. cathartica* or to a few species in the genus *Rhamnus* and should be considered in a future phase of the project. With the possible exception of *S. janiszewskae*, the few shoot borers and root borers associated with buckthorns in Europe lack host specificity at the genus level.

There are few genus-specific arthropods on *F. alnus*. Besides the leaf-hopper, *Z. suavis*, which was the most host-specific species found in this study, literature records indicate the existence of another five arthropod species known from *F. alnus* only. None of these were encountered during this study and additional surveys are needed to confirm their host ranges in the field in Europe. Current indications are that finding species-specific or genus-specific agents for biological control of *F. alnus* will be difficult.

Large numbers of herbivores are known to be associated with plants with larger geographical ranges (Lawton and Schroeder, 1977; Strong et al., 1984). *Rhamnus cathartica* and *F. alnus* have similar geographical distributions in Europe (Tutin, 1968), so range should not account for differences in the arthropod richness associated with the two

Table 3

Comparison of the frequency of occurrence of specialized buckthorn arthropods on *R. cathartica* and *F. alnus* in 59 allopatric and 20 sympatric sites (2002–05, Italy, Austria, Switzerland, Germany and the Czech Rep.)

Total No. of sites sampled:	Allopatric sites		Sympatric sites		
	32	27	20		
	<i>Rhamnus cathartica</i>	<i>Frangula alnus</i>	<i>Rhamnus cathartica</i>	<i>Frangula alnus</i>	<i>R. cathartica</i> and <i>F. alnus</i>
	Number of sites (%) with:		Number of sites (%) with:		
<i>Bucculatrix frangutella</i>	4 (12.5)	4 (14.8)	1 (5.0)	—	2 (10.0)
<i>Sorhagenia janiszewskae</i>	12 (37.5)	8 (29.6)	—	5 (25.0)	1 (5.0)
<i>Calybites quadrisignella</i>	1 (3.1)	—	2 (10.0)	—	—
<i>Stigmella catharticala</i>	1 (3.1)	—	1 (5.0)	—	—
<i>Stigmella rhamniella</i>	—	—	1 (5.0)	—	—
<i>Acrobasis romanella</i>	—	—	1 (5.0)	—	—
<i>Gonopteryx rhamni</i>	1 (3.1)	14 (51.9)	2 (10.0)	3 (15.0)	4 (20.0)
<i>Philereme vetulata</i>	6 (18.8)	—	8 (40.0)	—	—
<i>Philereme transversata</i>	3 (9.4)	—	5 (25.0)	—	—
<i>Triphosa dubitata</i>	17 (53.1)	—	5 (25.0)	—	—
<i>Ancylis apicella</i>	3 (9.4)	5 (18.5)	1 (5.0)	1 (5.0)	1 (5.0)
<i>Ancylis derasana</i>	3 (9.4)	2 (7.4)	2 (10.0)	—	1 (5.0)
<i>Cacopsylla rhamnicola</i>	4 (12.5)	—	1 (5.0)	—	—
<i>Trichochermes walkeri</i>	26 (81.3)	—	17 (85.0)	—	—
<i>Trioza rhamni</i>	23 (71.9)	—	13 (65.5)	—	—
<i>Zygina suavis</i>	—	3 (11.1)	—	2 (10.0)	—
<i>Oberea pedemontana</i>	—	—	—	—	2 (10.0)
<i>Phyllocoptes annulatus</i>	6 (18.8)	—	3 (15.0)	—	—

species. Centers of diversification of weed tribes or genera, which in turn are reflected by the highest number of congeneric or contribal species, are known to be the richest source of suitable herbivore species on certain weeds (Wapshere et al., 1989). Several studies found that taxonomic relatedness contributed significantly, though slightly, to arthropod species richness (e.g. Lawton and Schroeder, 1977; Neuvonen and Niemelä, 1981; Kennedy and Southwood, 1984). This is because related plants are likely to have the same chemical and physical traits and so are more likely to share herbivore insect species. Thus, plants with close relatives in a region (low taxonomic isolation) should have more herbivore species than those growing in isolation. In a study on the species richness of phytophagous insects and mites associated with 25 native tree genera in Germany, Brändle and Brandl (2001) concluded however that the importance of taxonomic isolation affecting herbivore species richness remains ambivalent. Contradictory results may be the consequence of the degree of taxonomic isolation (genus vs tribe or family), or the origin of the plant species considered (native vs introduced), the level of specificity of the herbivores considered (specialist vs generalist) or whether only a subset of the arthropod community is included in the study.

The genera *Rhamnus* and *Frangula* are predominant in the Old World and New World, respectively. In Europe there are 23 *Rhamnus* taxa as compared to four *Frangula* taxa. Thus, it appears that the evolution of *Rhamnus* and *Frangula* in isolation has led to specialization of arthropods on *Rhamnus* and *Frangula* species, with only a few specialist species on *F. alnus* in its native range in Europe and few species which are equally associated with both buckthorn species. Although we did not consider other sources of variability in the species richness of the arthropod fauna, such as plant abundance, this work supports the hypothesis that the species richness of specialized herbivores on native trees is affected by the taxonomic isolation of their hosts. It also shows that the search for, and selection of, potential biological control agents for a target weed can be facilitated by studying the arthropod species associated with related host plants from different lineages in centers of diversification.

Acknowledgments

We thank S. Izhovski, G. Fayvush, Ding Jianqing, A.M. Poswal, P. Baronio, N. Baldassari, A. Marini, H. Malicky, M. Tschirnhaus, C. Lambelet, R. Mayor, J.-P. Sorge, A. Bogenrieder, F. Schuhwerk, T. Le Bourgeois and A. Hampe for providing information on the distribution of buckthorn in Europe; S. Whitebread, B. Landry, J. Buszko, A. Haltrich, L.O. Hansen and R. Petanovic for providing information on arthropods associated with buckthorn; Z. Lastuvka, P. Lauterer, R. Bryner, M.J.W. Cock and M. Skuhrava for identifying most of the material collected on buckthorns; M. D'Alessandro for compiling literature data; E. Spiewak, B. Kiefer, T. Rheinhold, G.

Maia, A. Appleton, G. Cortat, and C. Péré for their field and laboratory assistance. We acknowledge M.J.M. Cock, A. McClay, R. DeClerck-Floate, K. Bolmgren and two anonymous reviewers for their most valuable comments on this manuscript. This project was funded by the United States Environmental Protection Agency—Great Lakes National Program Office, the Minnesota Environment and Natural Resources Trust Fund as recommended by the Legislative Commission on Minnesota Resources (LCMR), and by the Minnesota Department of Natural Resources.

References

- Amrine, J.W., Stasny, T.A.H., 1994. Catalog of the Eriophyoidea (Acarina: Prostigmata) of the world. Indira Publishing House, Michigan, USA, 804 pp.
- Baran, T., 1997. New record of *Sorhagenia lophyrella* (Douglas, 1846) (Lepidoptera: Cosmopterigidae) from Poland. *Wiadomosci-Entomologiczne* 16, 122 (in Polish).
- Barnes, H.F., 1951. Gall midges of economic importance. *Gall Midges of Trees*, vol. 5. Crosby Lockwood and Sons Ltd., London.
- Baronio, P., Marini, M., Sama, G., 1988. Studies on *Oberea pedemontana* Chevrolat 1856 (Coleoptera, Cerambycidae). *Monti e Boschi* 39, 45–52 (in Italian).
- Bergmann, A., 1952. Die Grossschmetterling Mitteleuropas. Jena, Urania Verlag GmbH.
- Bibby, T., 1983. Oviposition by the brimstone butterfly, *Gonepteryx rhamni* (L.), (Lepidoptera: Pieridae) in Monk's Wood, Cambridgeshire in 1982. *Entomologists' Gazette* 34, 229–234.
- Bittermann, J., 1997. Beitrag zur Kenntnis der Bionomie, Verbreitung und Gefährdung des Faulbaum-Glasflüglers *Synanthedon stomoxiformis* (Hübner, 1790). *Bayern Beiträge zur Bayerischen Entomofaunistik* 2, 131–139.
- Blackman, R.L., Eastop, V.F., 1994. Aphids on the World trees. An Identification and Information Guide. CAB International, Wallingford, 986 pp.
- Blaschke, P., 1914. Die Raupen Europas mit ihren Futterpflanzen. Grasers Verlag, Annaber Sachsen.
- Bolmgren, K., Oxelman, B., 2004. Generic limits in *Rhamnus* L. s.l. (Rhamnaceae) inferred from nuclear and chloroplast DNA sequence phylogenies. *Taxon* 53, 383–390.
- Brändle, M., Brandl, R., 2001. Species richness of insects and mites on trees: expanding Southwood. *Journal of Animal Ecology* 70, 491–504.
- Buhr, H., 1965. Bestimmungstabellen der Gallen (Zoo- und Phytoecidien) an Pflanzen Mittel und Nordeuropas. Band II. Veb Gustav Fischer Verlag, Jena.
- Buszko, J., 1992. Studies on the mining Lepidoptera of Poland. XII. Redescription of *Bucculatrix rhamniella* Herrich-Schäffer, 1855 (Bucculatricidae), with comments on its present distribution. *Polskie Pismo Entomologiczne* 61, 7.
- Catling, P.M., Porebski, Z.S., 1994. The history of invasion and current status of glossy buckthorn (*Rhamnus frangula*) in Southern Ontario. *Canadian Field Naturalist* 108, 305–310.
- Contarini, E., Garagnani, P., 1980. The cerambycids of the coastal pinewoods of *Ravenna* (Coleoptera). *Memorie della Societa Entomologica Italiana* 59, 49–57 (in Italian).
- Converse, C.K., 2001. Element Stewardship Abstract for *Rhamnus cathartica*, *Rhamnus frangula* (syn. *Frangula alnus*). The Nature Conservancy, Arlington, Virginia.
- Coulianos, C.-C., 1998. Annotated catalogue of the Hemiptera–Heteroptera of Norway. *Fauna Norvegica* 45, 11–40.
- de Freina, J.J., 1983. Studien ueber Biologie, Verbreitung, geographische Variabilitaet und Morphologie von *Gonepteryx farinose* (Zeller, 1847) nebst zusaetzlicher Erlauterung der Verbreitung und geographischen

- Variabilität von *Gonepteryx rhamni* (Linne, 1758) in Kleinasien. Mitteilungen der Münchner Entomologischen Gesellschaft 72, 9–55.
- de Freina, J.J., 1997. Die Bombyces und Sphinges der Westpalaearktis, Band 4, Sesiioidea: Sesiidae, EFW ed. Forschung & Wissenschaft, Verlag GmbH, München.
- Demelt, C. v., Franz, H., 1990. Ein systematisches Verzeichnis aller auf österreichischem Gebiet festgestellten Tierarten. Catalogus Faunae Austriae, vol. 15. Verlag der Oesterreichischen Akademie der Wissenschaft, Wien.
- Doczkal, D., Rennwald, E., 1992. Observations on the habits, distribution, and endangerment of the clearwing moth *Synanthedon stomoxiformis* (Huebner, 1790) in Baden-Wuerttemberg (Lepidoptera: Sesiidae). Atalanta Marktleuthen 23 (1–2), 259–274 (in German).
- Emmet, A.M., 1969. Discovery of the larva of *Sorhagenia rhamniella* Zeller. The Entomologist's Record 81, 20–21.
- Forster, W., Wohlfahrt, T.A., 1981. Die Schmetterlinge Mitteleuropas; Band 5: Spanner (Geometridae), Franckische Verlagshandlung, Stuttgart, 312 pp.
- Fox, L.R., Morrow, P.A., 1981. Specialization: species property or local phenomenon? Science, 887–893.
- Frisch, J., 1992. Ein neuer Bockkäfer für die Fauna Kärntens—*Oberea pedemontana* Chevrolat 1856 (Coleoptera, Cerambycidae). Carinthia II 182, 637–642.
- Frohawke, F.W., 1940. Food-plants of *Gonepteryx rhamni*. The Entomologist 73, 68–69.
- Gale, S.W., 2001. Control of the invasive exotic *Rhamnus cathartica* in temperate North America forests. <<http://www.hort.agri.umn.edu/h5015/00papers/gale.htm>>.
- Gassmann, A., Tosevski, I., Appleton, A., Cortat, G., Péré, C., Reinhold, T., 2006. Biological Control of Buckthorns (*Rhamnus cathartica* and *Frangula alnus*). Report 2004–05, CABI Bioscience, Switzerland Centre, 62 pp.
- Gollner-Scheiding, U., 1972. Beitrag zur Heteropteren-Fauna Brandenburg. 2. Übersicht über die Heteropteren von Brandenburg. Veröffentl. Veröffentl. Bizirkshemat Museum Potsdam (25/26), 5–39.
- Gourley, L.C., 1985. A study of the ecology and spread of buckthorn (*Rhamnus cathartica* L.) with particular reference to the University of Wisconsin arboretum. Department of Landscape Architecture, University of Wisconsin, Madison, Wisconsin, USA.
- Grubov, V.I., 1949. Monography of *Rhamnus* L. s.l. In: Schischkin, B.K. (Ed.), Flora et Systematica Plantae Vasculares. Academiae Scientiarum USSR, Leningrad, pp. 14–425.
- Gutierrez, D., Thomas, C.D., 2000. Marginal range expansion in a host-limited butterfly species *Gonepteryx rhamni*. Ecological Entomology 25, 165–170.
- Haber, E., 1997. Invasive Exotic Plants of Canada. Fact sheet No. 6. Glossy Buckthorn—*Frangula alnus* P. Mill. <<http://info-web.magi.com/~ehaber/factcbck.html>>, National Botanical Services, Ottawa, ON, Canada.
- Hampe, A., Arroyo, J., Jordano, P., Petit, R.J., 2003. Rangewide phylogeography of a bird-dispersed Eurasian shrub: contrasting Mediterranean and temperate glacial refugia. Molecular Ecology 12, 3415–3426.
- Hannemann, H.-J., 1964. Kleinschmetterlinge oder Microlepidoptera. II. Die Wickler (s. l.) (Cochylidae und Carposinidae), die Zünselartigen (Pyraloidea). In: Dahl, F., Dahl, M., Peus, F. (Eds.), Die Tierwelt Deutschlands und der angrenzenden Meeressteile. 50. Teil. Gustav Fischer Verlag, Jena.
- Harder, D.E., Chong, J., 1983. Virulence and distribution of *Puccinia coronata* in Canada in 1982. Canadian Journal of Plant Pathology 5, 185–198.
- Heath, J. (Ed.), 1976. Moths and butterflies of Great Britain and Ireland. Vol. 1: Micropterygidae–Heliozelidae. Springer, Berlin.
- Heath, J., Emmet, A.M., 1985. The Moths & Butterflies of Great Britain and Ireland. Vol. 2: Cossidae–Heliodinidae. Harley Books, Colchester, UK.
- Heath, J., Emmet, A.M., 1989. The Moths & Butterflies of Great Britain and Ireland. Vol. 7: Hesperidae–Nymphalidae. Harley Books, Colchester, UK.
- Heidorn, R., 1991. Vegetation management guideline: exotic buckthorns—common buckthorn (*Rhamnus cathartica* L.), glossy buckthorn (*Rhamnus frangula* L.), *Dahurian buckthorn* (*Rhamnus davurica* Pall.). Natural Areas Journal 11, 216–217.
- Heie, O.E., 1986. The Aphidoidea (Hemiptera) of Fennoscandia and Denmark. III. Family Aphididae: subfamily Pterocommatinae & tribe Aphidini of subfamily Aphidinae. Fauna Entomologica Scandinavica, vol. 17. E.J. Brill/Scandinavian Science Press, Leiden, 314 pp.
- Hering, E.M., 1957. Bestimmungstabellen der Blattminen von Europa. Den Haag, Junk.
- Horion, A.D., 1974. Faunistik der Mitteleuropäischen Käfer. Cerambycidae. Bodensee, Ueberlingen.
- Houard, C., 1909. Les Zoocécidies des Plantes d'Europe et du Bassin de la Méditerranée. Librairie Scientifique A. Hermann et fils, Paris.
- Ivinskis, P.P., Ionaitis, V., Petrauskas, V., 1982. 138 species of Lepidoptera new to the Lithuanian SSR, collected from 1968 to 1982. Novye-izredkie-dlya-Litovskoi-SSR-vidy-nasekomykh, pp. 28–47 (in Russian).
- Jacobi, K., Menne, B., 1991. Reproductive biology of the troglomorphic looper moth *Triphosa dubitata* L. (Lepidoptera, Geometridae). Carolinia 49, 129–130.
- Johnston, M.C., Johnston, L.V., 1978. *Rhamnus*. In: Flora Neotropica Monograph 20. New York Botanical Garden, New York, pp. 1–96.
- Kennedy, C.E.J., Southwood, T.R.E., 1984. The number of species of insects associated with British trees: a re-analysis. Journal of Animal Ecology 53, 455–478.
- Knight, K.S., Kurylo, J.S., Endress, A.G., Stewart, J.R., Reich, P.B., 2007. Ecology and ecosystem impacts of common buckthorn (*Rhamnus cathartica*): a review. Biological Invasions 9, 925–937.
- Lawton, J.H., Schroeder, D., 1977. Effects of plant type, size of geographical range and taxonomic isolation on number of insect species associated with British plants. Nature 265, 137–140.
- Lekic, M., Mihajlovic, L., 1976. A contribution to the investigation of the ecology of *Oberea pedemontana* Chevrolat on *Rhamnus cathartica* as a host plant (in Serbian). Glasnik Sumarskog Fakulteta Serbia 50, 67–72.
- Leonard, K.J., 2003. Regional frequencies of virulence in oat crown rust in the United States from 1990 through 2000. Plant Disease 87, 1301–1310.
- Linnaeus, C. von, 1753. Species Plantarum. Holmiae, Impensis L. Salvii, vol. 1, pp. 193–195.
- Louda, S.M., Kendall, D., Connor, J., Simberloff, D., 1997. Ecological effects of an insect introduced for the biological control of weeds. Science 277, 1088–1090.
- Malicky, H., Sobhian, R., Zwölfer, H., 1970. Investigations on the possibilities of biological control of *Rhamnus cathartica* L. in Canada: Host ranges, feeding sites, and phenology of insects associated with European Rhamnaceae. Zeitschrift für angewandte Entomologie 65, 77–97.
- McKay, H.V., 1991. Egg-laying requirements of woodland butterflies; Brimstones (*Gonepteryx rhamni*) and alder buckthorn (*Frangula alnus*). Journal of Applied Ecology 28, 731–743.
- McLean, I.F.G., 1993. The host plant associations and life history of *Trichochermes walkeri* Förster (Psylloidea: Trioziidae). British Journal of Entomology and Natural History 6, 4.
- Medan, D., 1994. Reproductive biology of *Frangula alnus* (Rhamnaceae) in southern Spain. Plant Systematics and Evolution 193, 173–186.
- Meyer, J., 1987. Plant Galls and Gall Inducers. Gebrüder Bornträger, Berlin, Stuttgart.
- Michalska, Z., 1996. Nepticulidae (Lepidoptera) of Roztocze. Srodkowe Fragmenta Faunistica Warsaw 39, 61–69.
- Mihajlovic, L., 1978. First contribution to the knowledge of the superfamily Pyraloidea. Fauna of Serbia 2, 177–201.
- Miller, P., 1754/1768. The Gardener's Dictionary, fourth ed., Bd. 1–3, Rivington, London (8th ed., published in 1768).
- Neuvonen, S., Niemelä, P., 1981. Species richness of macrolepidoptera on Finnish deciduous trees and shrubs. Oecologia 51, 364–370.
- Okopnyi, N.S., Poddubnyi, A.G., 1983. Mechanism of gall formation on leaves of buckthorn infected with psyllid larvae *Rhamnus cathartica*

- attacked by *Trichohermes walkeri*. Dokl. Bot. Sci. Akad. Nauk. SSSR (265/267), 114–117.
- Ossiannilsson, F., 1981. The Auchenorrhyncha (Homoptera) of Fennoscandia and Denmark. Vol. 7, Part 2: The families Cicadidae, Cercopidae, Membracidae, and Cicadellidae (excl. Deltocephalinae), Scandinavian Science Press Ltd., pp. 584–586.
- Ossiannilsson, F., 1992. The Psylloidea (Homoptera) of Fennoscandia and Denmark. Fauna Entomologica Scandinavica, 26.
- Pemberton, R.W., 2000. Predictable risk to native plants in weed biological control. *Oecologia* 125, 489–494.
- Pollard, E., Hall, M.L., 1980. Possible movement of *Gonepteryx rhamni* (L.) (Lepidoptera: Pieridae) between hibernating and breeding areas. *Entomologist's Gazette* 31, 217–220.
- Puplesis, R., 1994. The Nepticulidae of Eastern Europe and Asia: Western, Central and Eastern Parts. Leiden. Backhuys Publishers, The Netherlands.
- Puplysis, R.K., 1984. Contribution to the knowledge of pigmy moths of the genus *Stigmella* Schrank (Lepidoptera, Nepticulidae) of Lithuania (in Russian). *Acta Entomologica Lituanica* 7, 72–85.
- Rameau, J.C., Mansion, D., Dumé, G., Timbal, J., Lecointe, A., Dupont, P., Keller, R., 1989. Flore forestière française. Guide écologique illustré. 1. Plaines et collines. Institut pour le Développement Forestier, Ministère de l'Agriculture et de la Forêt.
- Randall, J.M., Marnelli, J., 1996. Invasive plants: weeds of the global garden. Brooklyn Botanic Gardens, Inc., Brooklyn, NY, 111 pp.
- Razowski, J., 1970. Cochylidae. In: Amsel, H.-G., Gregor, F., Reisser, H. (Eds.), *Microlepidoptera Palaearctica*, vol. 3. Verlag Georg Fromme & Co, Wien.
- Razowski, J., 2003. Tortricidae (Lepidoptera) of Europe. Vol. 2. Olethreutinae, Frantisek Slamka, Bratislava, 301 pp.
- Ripley, I., 1984. The purple hairstreak and brimstone butterflies in Northern Ireland. *Entomologist's Gazette* 35, 5.
- Scamoni, A., 1985. Ueber das ökologische und soziologische Verhalten von *Rhamnus cathartica* L. und *Frangula alnus* Mill. in der planaren Stufe der DDR. *Feddes Repertorium* 96, 139–144 (in German).
- Schmidt, K.A., Whelan, C.J., 1999. Effects of exotic *Lonicera* and *Rhamnus* on songbird nest predation. *Conservation Biology* 13, 1502–1506.
- Skinner, B., 1984. Colour Identification Guide to Moths of the British Isles (Macrolepidoptera), Viking.
- Spatenka, K., Gorbunov, O., Lastuvka, Z., Tosevski, I., Arita, Y., 1999. Handbook of Palaearctic Macrolepidoptera. Sesiidae—Clearwing Moths, vol. 1. Gem Publishing Company, Wallingford, England.
- Speight, M.C.D., Cogan, B.H., 1979. *Agrometopia wahlbergi*, *Clusiodes caledonica* and *Stigmella catharticea*: insects new to Ireland. *Irish Naturalists' Journal* 19, 401–402.
- Stadie, D., 1995. Lebensweise und Verbreitung des Kreuzdornglasflüglers *Synanthedon stomoxiformis* (Hübner, 1790) in Thüringen und Sachsen-Anhalt (Lep., Sesiidae). *Entomologische Nachrichten und Berichte* 39, 219–223.
- Stelter, H., 1975. Untersuchungen über Gallmücken XVIII. *Wachtliella krumbholzi* n. sp. in Fruchtgallen von *Rhamnus cathartica* L. (in German). *Deutsche Entomologische Zeitschrift* 22, 157–162.
- Strong, D.R., Lawton, J.H., Southwood, R., 1984. Insects on plants. Community Patterns and Mechanisms. Blackwell Scientific Publications, Oxford, 313 pp.
- Tutin, T.G., 1968. Rhamnaceae. In: Tutin, T.G., Heywood, V.H., Burges, N.A., Moore, D.M., Valentine, D.H., Walters, S.M., Wegg, D.A. (Eds.), *Flora Europaea*, vol. 2. Cambridge University Press, Cambridge, MA, pp. 243–245.
- USDA/NRCS, 2001. The PLANTS Database, Version 3.1. <<http://plants.usda.gov>>, National Plant Data Center, Baton Rouge, LA 70874-4490, USA.
- Voegtlin, D.J., O'Neil, R.J., Graves, W.R., Lagos, D., Yoo, H.J.S., 2005. Potential winter hosts of soybean aphid. *Annals of the Entomological Society of America* 98, 690–693.
- Wapshere, A.J., Delfosse, E.S., Cullen, J.M., 1989. Recent development in biological control of weeds. *Crop Protection* 8, 227–250.
- Zerova, M.D., Mamontova, V.A., Ermolenko, V.M., Djakonchuk, L.A., Sinev, C.J., Kozlov, M.V., 1991. Neseconomie-Galloobrazovateli kulturnih i dekorastujusih rastenii evropeiskoi casti SSSR., Kiev Naukova Dumka.

Population Biology of Garlic Mustard (*Alliaria petiolata*) in Minnesota Hardwood Forests

Laura C. Van Riper, Roger L. Becker, and Luke C. Skinner*

Garlic mustard, a biennial forb native to Europe, has invaded native ecosystems in forested regions in the United States. In anticipation of a biological control program being implemented in the United States for this plant, a garlic mustard monitoring program was initiated. The objective of this study was to characterize garlic mustard populations and the associated plant communities and their response to environmental conditions in Minnesota hardwood forest ecosystems. Additionally, we developed a baseline for long-term studies to determine future benefits and impacts of biological control agents on plant communities infested with garlic mustard, should they be released. To monitor garlic mustard populations, we used a nationally standardized protocol in which data were collected on garlic mustard population density and cover, garlic mustard plant heights and silique production, insect damage to garlic mustard, cover of the associated plant community, and litter cover. We also collected data on available photosynthetically active radiation in the understory. The results underscore the variability in garlic mustard population dynamics. At only 6 of 12 sites did garlic mustard densities follow the predicted two-point cycles due to their biennial life cycle, with the first- or second-year life stage dominating in any given year. Available light did not differ strongly among sites, but shading by adult plants is implicated in keeping the populations of first-year plants low. Sites with greater garlic mustard cover had lower native species richness and cover than sites with lower garlic mustard cover. Absent biological control agents, garlic mustard is currently experiencing very little herbivory in Minnesota with an average of 2% of leaf area removed by herbivores. Our work shows the importance of pre-release monitoring at multiple sites over multiple years to adequately characterize populations. Without control, garlic mustard will likely continue to have negative impacts on northern forests.

Nomenclature: Garlic mustard, *Alliaria petiolata* (Bieb.) Cavara & Grande.

Key words: Pre-release monitoring, invasive species, biological control, herbivory, population cycling, biennial.

Garlic mustard [*Alliaria petiolata* (Bieb.) Cavara & Grande] is an invasive, nonnative species that is invading forested regions throughout the United States (Cavers et al. 1979; Meekins et al. 2001; Rodgers et al. 2008). Garlic mustard is a concern because of its ability to invade relatively undisturbed, diverse forests; form dense populations; and decrease abundance of native species (Blossey et al. 2001; Nuzzo 1999). Garlic mustard is part of a complex community, and how it interacts with members of that community is not clear. Abundant native deer and

nonnative earthworms are likely facilitating garlic mustard invasion (Eschtruth and Battles 2009a; Nuzzo et al. 2009). Nonnative earthworms have invaded many forests and denuded the litter layer, altered soil processes, and decreased native species abundance (Bohlen et al. 2004; Hale et al. 2005). Bartuszevige et al. (2007) found that garlic mustard seedlings had the greatest establishment in plots with litter removed vs. control or litter added plots. Blossey et al. (2005) observed that garlic mustard was often found in areas with little to no litter layer, indicating that garlic mustard may succeed in sites that have been invaded by earthworms. They observed that the overpopulation of deer in many areas has put additional pressure on native plants, noting that garlic mustard was generally grazed less than native plants and so may have done better than natives in sites with high deer population density.

High light availability can increase the biomass and seed production of garlic mustard plants (Meekins and

DOI: 10.1614/IPSM-D-09-00008.1

*First and second authors: Post-Doctoral Research Associate and Professor, Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108; and third author: Supervisor, Invasive Species Unit, Minnesota Department of Natural Resources, St. Paul, MN 55155. Corresponding author's E-mail: scho0536@umn.edu

Interpretive Summary

Invasive garlic mustard (*Alliaria petiolata*) has become abundant in many forested regions of the United States. This study examined the fluctuations of garlic mustard populations over time and their relationship with native species, levels of leaf litter, photosynthetic radiation, and insect herbivores. At half of the 12 monitoring sites, garlic mustard populations showed strong two-point cycling with alternating dominance of the first- and second-year life stages. Increased garlic mustard cover was negatively correlated with native species richness and cover. All sites had litter layers that had been significantly impacted by earthworms. Light was a key factor in understanding garlic mustard populations. Adult plant cover is higher where light is more abundant, but high cover of adult plants produces shade and can cause low cover of seedling plants. We found that less than 2% of garlic mustard leaf area is currently being damaged by herbivores in Minnesota.

These results have implications for both the release of potential biological control agents and restoration of garlic mustard invaded sites. The current lack of strong herbivory indicates that garlic mustard populations may be highly impacted by the release of biological control insects. If insects are to be released, it is useful to monitor the site for several years prior to release to determine the current densities of garlic mustard and normal fluctuations in population. If a site is strongly cyclical, it may be beneficial to time the release of biological control insects to match the life stage of the insect with the correct life stage of garlic mustard. When working to restore a site that has been heavily invaded by garlic mustard, the level of earthworm impact, the number and abundance of native species that remain, and any changes to the light available from the canopy should all be considered as factors that could influence the recovery of the site, in addition to the potential decrease in garlic mustard.

McCarthy 2000; Myers et al. 2005), ultimately influencing garlic mustard population size. Site differences in garlic mustard cover may be due to the amount of light a site receives (Eschtruth and Battles 2009b). Adult garlic mustard plants can shade out the seedlings growing beneath them, causing one life stage to dominate the other.

Presence of garlic mustard may change soil properties in forests. Garlic mustard has allelopathic root exudates that can inhibit germination in some species (Prati and Bossdorf 2004). The root exudates have also been found to have a negative impact on mycorrhizal fungi (Stinson et al. 2006; Wolfe et al. 2008). Many late-successional native species are dependent on mycorrhizae, so the loss of mycorrhizae can negatively affect native species abundance.

Due to the number of negative impacts of garlic mustard, a program was initiated to develop biological control agents for North America (Blossey et al. 2001). Currently, three weevil species are being tested at the University of Minnesota quarantine facility in cooperation with CABI Europe-Switzerland to determine their host specificity and suitability as biological control agents. The three species are the root-crown feeding weevil *Ceutorhynchus scrobicollis* Nerensheimer and Wagner and the stem-mining weevils *Ceutorhynchus alliariae* Brisout and

Ceutorhynchus robertii Gyllenhal (Blossey et al. 2001; Katovich et al. 2005). It is anticipated that the release of biological control agents will cause a reduction in the cover, density, height, and reproductive output of garlic mustard plants and an increase in the amount of visible leaf damage. A petition to approve the release of *C. scrobicollis* was submitted to the United States Department of Agriculture (USDA)-Animal and Plant Health Inspection Service (APHIS) Technical Advisory Group in 2008. In 2009, the Technical Advisory Group recommended host-specificity testing for a few additional key plant species prior to approval. The additional tests are currently underway, and completion is expected in early 2010.

Historically, the benefits and impacts of biological control agents on plant communities have been difficult to assess due to the lack of pre-release baseline data (Blossey 1999). Garlic mustard is a biennial species whose population densities can fluctuate significantly from year to year (Meekins and McCarthy 2002) requiring several years of plant monitoring data to characterize invaded plant communities. By monitoring other plant populations growing with garlic mustard, we can gain a better understanding of the relationship and impacts of garlic mustard on native and nonnative plant species. The objective of this study was to characterize garlic mustard populations and the associated plant communities and their response to environmental conditions in Minnesota hardwood forest ecosystems. These data are a baseline for long-term studies from which to determine future benefits and impacts of biological control agents—should they be released—on plant communities infested with garlic mustard. By monitoring garlic mustard populations, we have a greater understanding of the role of density-dependent population dynamics, and the relationship between garlic mustard and factors such as light, litter cover, and native species.

Materials and Methods

Twelve monitoring study sites were established in hardwood forests in Minnesota. The sites are located between 44°6.600' and 45°19.356' N, and 92°3.821' and 94°56.667' W. To monitor these plant communities, we used the standardized protocol developed by the Ecology and Management of Invasive Plants Program in 2003 (available at <http://www.invasiveplants.net>). Sites were selected according to the following criteria: (1) the managers of the site agreed to suspend all management that could impact garlic mustard plants and biological control insects in the area (no herbicide treatments, burns, or pulling of garlic mustard populations), (2) the site had a well-established garlic mustard population of sufficient size to hold four 50-m (164-ft) transects spaced at least 10 m apart, and (3) the site could not be subject to flooding,

Table 1. Garlic mustard monitoring sites in Minnesota, 2005 to 2008. The identification (ID) column lists the abbreviation for that site as found in the figures.

Site no.	ID	Site name	City	County	Habitat type	Latitude, Longitude
1	BP	Baker Park Preserve*	Maple Plain	Hennepin	Upland	45°02.427', 93°37.195'
2	CR	Coon Rapids Dam Regional Park	Coon Rapids	Anoka	Floodplain	45°07.975', 93°17.841'
3	CG	Cottage Grove Ravine Regional Park	Cottage Grove	Washington	Upland	44°48.480', 92°53.960'
4	FS	Fort Snelling State Park*	Saint Paul	Ramsey	Floodplain	44°52.373', 93°11.634'
5	HP	Hilloway Park	Minnetonka	Hennepin	Upland	44°57.552', 93°26.098'
6	LL	Luce Line	Long Lake	Hennepin	Upland	44°58.441', 93°35.137'
7	NE	Nerstrand State Park, Prairie Creek SNA*	Nerstrand	Rice	Upland	44°21.527', 93°05.809'
8	PB	Pine Bend Bluffs SNA*	Inver Grove Heights	Dakota	Upland	44°47.076', 93°01.732'
9	PL	Plainview – private land	Plainview	Winona	Upland	44°06.600', 92°03.821'
10	WN	Warner Nature Center*	Marine on St. Croix	Washington	Upland	45°10.853', 92°49.641'
11	WH	Westwood Hills Nature Center	St. Louis Park	Hennepin	Upland	44°58.301', 93°23.692'
12	WI	Willmar - private land	Willmar	Kandiyohi	Upland	45°19.356', 94°59.667'

* = one of five sites established in time for spring 2005 data collection.

which might drown biological control insects and prevent the establishment of insect populations when released. At each site, 20 permanent 1-m by 0.5-m plots (0.5-m² quadrats) were established. The plots were placed along four 50-m transects with each transect containing five plots spaced 10 m from each other. Transects were placed through the center of the infestation, and each transect was at least 10 m from the nearest transect. According to the protocol, all plots must initially contain garlic mustard since the purpose is to monitor the impacts of biological control agents on garlic mustard, rather than monitor spread of garlic mustard populations. If there were no garlic mustard plants in a specific plot location along the transect 10 m from the previous plot, that plot was shifted down the transect to the next garlic mustard plant.

In June 2005, the initial five garlic mustard monitoring sites were established and data were collected. An additional 7 sites were established by the fall monitoring data collection period in October 2005 for a total of 12 sites (Table 1). In 2006 and 2007, data were collected from all 12 sites in June and October. All sites are upland deciduous forests, except for Coon Rapids (CR) and Fort

Snelling (FS), which are floodplain forests, although flooding is rare due to management of adjacent rivers. The most common canopy tree species were *Acer negundo* L. (box elder), *Acer saccharum* Marsh. (sugar maple), *Fraxinus pennsylvanica* Marsh. (green ash), *Quercus rubra* L. (northern red oak), *Ulmus rubra* Muhl. (slippery elm), and *Tilia americana* L. (basswood). The most common understory tree species are *A. negundo*, *A. saccharum*, *Celtis occidentalis* L. (hackberry), *Ostrya virginiana* (P. Mill.) K. Koch (ironwood), *Rhamnus cathartica* L. (common buckthorn), and *U. rubra*.

Following the standard protocol, monitoring data were collected in June and October from 2005 to 2008. Garlic mustard is a biennial, and the data collected reflects the various life stages. Seedlings emerge in the spring, and by the fall of that year, develop into basal rosettes. Rosettes overwinter and bolt in the spring of the second year forming adult flowering stalks. Adult plants flower in the spring, set seed in the summer, and senesce by the fall. Spring data include both the first-year seedling and second-year adult stages, while only the first-year rosette stage is present in the fall. The adult plants can be further divided

into those with siliques present and those with no siliques present. In each plot, all first-year garlic mustard plants were counted and a visual estimate of their percent cover made. In the spring, each second-year garlic mustard plant in each plot was measured to determine its height. The number of siliques of that individual was counted. The list of each adult plant height and number of siliques also resulted in a count of all adult plants in each plot. Visual estimates of percent cover of second-year garlic mustard plants were also made. Each year, the second-year plants senesced by October, so no fall data were collected on second-year plants.

In each plot, all other species present were identified and visual percent cover estimated for each species. Plants were identified to species when possible, and their percent cover in the plot recorded. For analysis, species were categorized as native, nonnative, or unknown (species that could not be identified to the taxonomic level where native or nonnative status could be determined). The number of native and nonnative species per quadrat was determined, and average native and nonnative species richness per 0.5-m² quadrat for each site was calculated. All native or nonnative determinations were based on the Minnesota Department of Natural Resources species list for Minnesota (2009).

Data were also collected to characterize herbivore damage to garlic mustard. Within each plot, a visual estimate was made of the percent of garlic mustard leaf area that was removed due to insect feeding. The following types of damage on garlic mustard plants within the plot were noted when present: leaf miner damage, windowpane feeding, edge feeding, holes, spittle bug presence, scale, browse, or disease.

Litter depth and ground cover at the site were also characterized. In the center of each half of each plot, a ruler was used to measure litter depth to the nearest cm. These two readings were then averaged to give a mean litter depth for each plot. For each plot, visual estimates were made of the percent cover of the ground that was composed of bare soil, wood, leaf litter, or rock. Low litter depths, low percent cover of leaf litter, and high percent cover of bare soil are indicative of sites that have high levels of disturbance by invasive earthworms (Bohlen 2004; Hale et al. 2005).

The amount of photosynthetically active radiation (PAR, 400 to 700 nm) penetrating the canopy was measured to determine if light differed among the sites and plots. Measurements were taken within 2 hr of solar noon, between August 11 and September 9, 2008. Tree leaves had not begun to change color or senesce. A LI-190SA point quantum light sensor with an LI-1000 data logger¹ was used to measure PAR levels in an area of full sun. The data logger sampled PAR levels every 5 s and recorded the average PAR level for 1-min intervals. Concomitantly, a 1-m LI-191SA line quantum light sensor and an LI-189

visual display² were used to take PAR measurements in plots under the forest canopy, placing the line sensor along the center of the 1-m-long axis of each plot. The PAR readings and time of each reading were recorded at 1 m above the soil surface to measure PAR transmitted through the forest canopy and available at the top of adult garlic mustard canopies, and at the soil surface to measure PAR available to seedling and rosette garlic mustard plants. The percent of full sun PAR incident at these two levels in the canopy was determined by dividing by the PAR reading under the canopy by the PAR reading in full sun at the time of the plot reading.

Data were summarized by season and site. In all graphs, the error bars are standard errors. A one-way ANOVA was used to determine if the percent of PAR reaching 1 m or the soil surface differed ($P = 0.05$) among the sites (Oehlert and Bingham 2005). Regressions and correlations were used to determine the strength of relationships among variables (Statistix 7 2000).

Results and Discussion

Fluctuations in Garlic Mustard Populations Over Time.

Garlic mustard's biennial life cycle added to the complexity of garlic mustard cover and population density over time. At some sites, one life stage clearly dominated in each year, while other sites had populations that were stable, increasing, or decreasing. Figure 1 is a pictorial example of life cycle dominance at Baker Park (BP), a site dominated by adult flowering plants in spring 2005 with few seedlings present that resulted in few rosettes by the fall of 2005. In the spring of 2006, the seedling stage dominated with many seedlings and very few adults. By fall 2006, rosettes were the dominant growth stage. Half of the sites studied (6 of 12) showed this pattern of one life stage dominating in a given year, but at each of these 6 sites, both first- or second-year life stages were present in a given year (Figure 2a). Two sites, Warner Nature Center (WN) and Westwood Hills (WH) showed strong cycling in all 4 yr with rosette densities peaking every other year, while four other sites showed cycling in only 3 of the 4 yr (CR, CG, LL, and NE). Populations were relatively stable at 2 of the 12 sites, FS and Hilloway Park (HP), with little variation in rosette population density from year to year with first- and second-year plants coexisting each year. Population densities increased at Willmar (WI) and decreased at Pine Bend (PB) from 2005 to 2008.

Patterns that were clear in the fall data were less visible in the spring, possibly an artifact of fewer replications included in the spring 2005 data. In most cases, the sites that showed strong year to year population cycling of rosettes in the fall also showed strong cycling of spring population densities of seedlings and adults (Figure 2b, c). For example, the population density of adult plants at WN

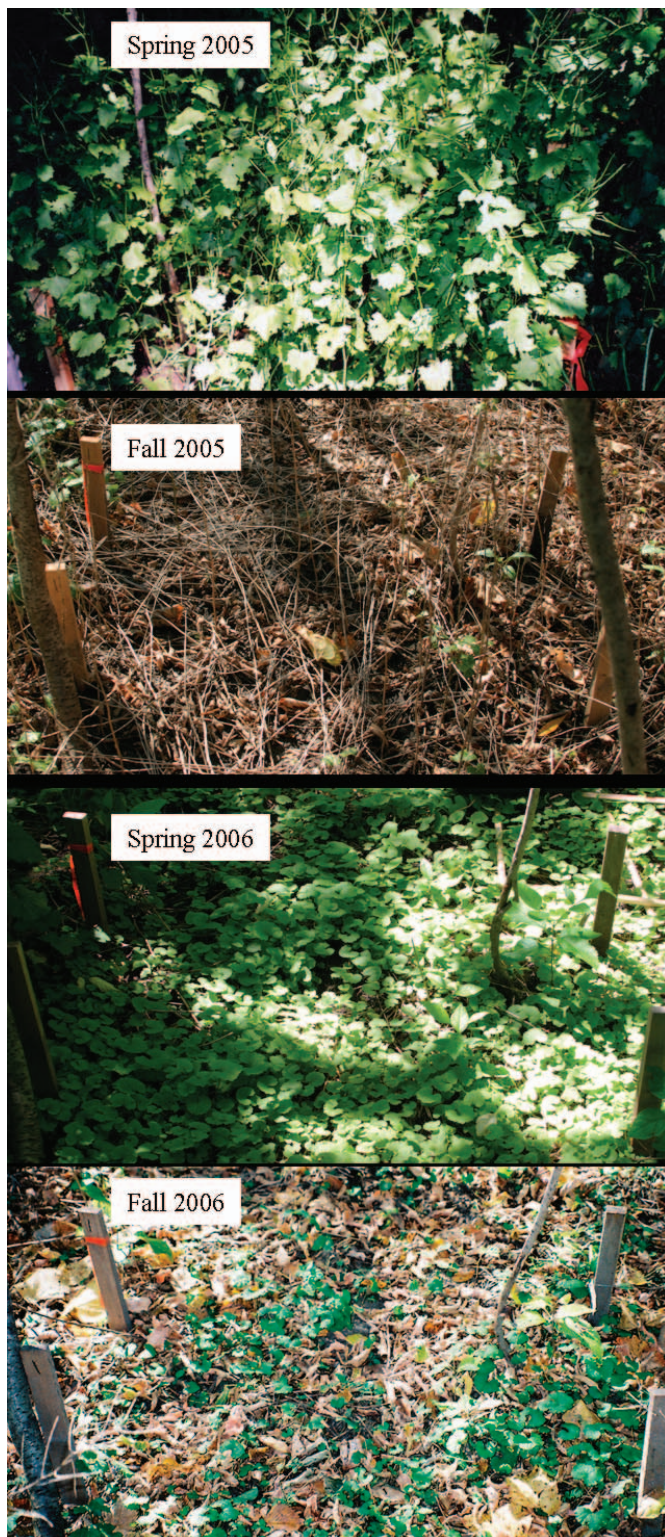


Figure 1. Photos of a single plot over time, showing a typical biennial cycle of the dominance of different garlic mustard life stages at Baker Park, MN, 2005 to 2006. The adult flowering plants were dominant in spring 2005 with few seedlings present. By fall 2005, the adults senesced and there was little other vegetation present. In spring 2006, there was a carpet of garlic

fluctuated widely, going from an average of 1 plant m^{-2} in 2005 to 85.2 plants m^{-2} in 2006 and then down to 15.4 plants m^{-2} in 2007 (Figure 2c). The low of 1 plant m^{-2} (in 2005) at WN and the high of 133 plants m^{-2} (in 2008) at WN were the lowest and highest adult garlic mustard population densities found across all sites and years. Seedling densities also ranged widely, from a low of 8.2 plants m^{-2} to a high of 301.2 plants m^{-2} on average at CR (Figure 2b).

High mortality occurred as garlic mustard plants progressed through life stages. Many seedlings died before reaching the fall rosette stage. The mean May to October mortality for garlic mustard seedlings averaged across all sites was 47, 52, and 77% in 2006, 2007, and 2008, respectively. Winter survival fluctuated dramatically across years. The mean mortality of rosettes overwintering to form adult plants averaged across sites was 7, 45, and 18% for 2005 to 2006, 2006 to 2007, and 2007 to 2008, respectively. Year to year differences indicate that factors such as precipitation and temperature in a given year can strongly influence garlic mustard survival. For example, the high mortality for first-year garlic mustard plants in 2008 may have been due to low precipitation in that year: 37 cm of precipitation from May to October 2008 vs. the 30-yr (1971 to 2000) average of 52 cm (Minnesota Climatology Working Group 2009).

In addition to monitoring garlic mustard population densities, we also monitored the percent cover of garlic mustard. At some sites, small numbers of plants were large and covered a large percentage of the plot, while at other sites, large numbers of very small plants covered only a small area. The average total percent cover of garlic mustard in the spring (adults plus seedlings) ranged from 20 to 70% among the sites (Figure 3). Garlic mustard cover decreased in the fall because the adult plants had set seed and senesced, so only first-year rosettes were present. Average total garlic mustard cover (seedlings plus adults in the spring, rosettes in the fall) did vary from year to year, although the range of garlic mustard cover was similar from year to year.

When garlic mustard populations are governed by density-dependence, they show a two-point cycle with the adult and seedling/rosette stages alternating in dominance (Pardini et al. 2009). In our study, only half of the sites show this two-point cycle. The population dynamics at the other six sites may not have been governed by density-dependence. Populations of garlic mustard in poor habitats with low population densities do not exhibit the two-point cycle, and high density populations are more likely to cycle (Pardini et al. 2009). However, in our study, the six sites

←
mustard seedlings. By fall 2006, the surviving seedlings had grown into rosettes.

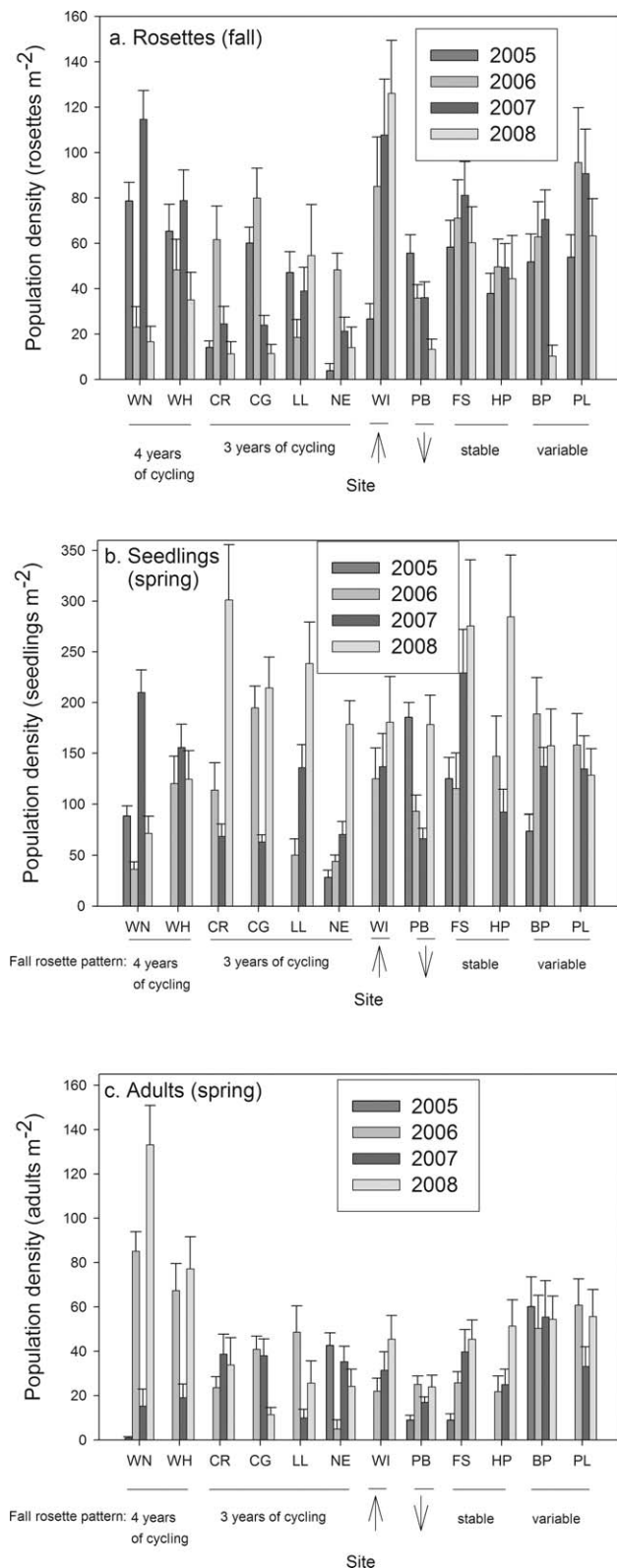


Figure 2. Mean population density (\pm standard error [SE]) of (a) garlic mustard rosettes, (b) seedlings, and (c) adults over time at 12 monitoring sites in Minnesota, 2005 to 2008. Data were collected on the rosettes in the fall at all 12 sites for 2005 to

without cycling did not have lower population densities than the sites where cycling occurred. Garlic mustard populations can vary greatly from year to year and with moderate levels of mortality can fall into complex and chaotic population dynamics (Pardini et al. 2009). Intraspecific competition favors the separation of the two age classes (Winterer et al. 2005). Dense garlic mustard itself can suppress its own seedlings, so sites that have a rapid establishment of garlic mustard may show cycling as a legacy of the initial wave of garlic mustard plants (Meekins and McCarthy 2002).

Within a larger population, there can be subpopulations that are established by different colonization events, and these subpopulations may cycle out of synch with one another (Meekins and McCarthy 2002). It is possible that there was subpopulation cycling at the non-cycling monitoring sites but that, if present, occurred at a smaller scale than we analyzed. For example, half the plots could have been dominated by seedlings, while the other half were dominated by adults, and together they even out. To see if individual plots tended to be dominated by one life stage or another, we analyzed data from individual plots from the six non-cycling sites. The total amount of spring garlic mustard cover (seedling cover + adult cover) was determined for the spring evaluations and the portion of that cover that was accounted for by each life stage determined. In 2006, 40% of the individual plots at the six non-cycling sites were dominated by seedling garlic mustard plants (seedlings accounting for 80 to 100% of garlic mustard cover, with the remaining 0 to 20% of garlic mustard cover comprised of adult plants). The reverse occurred in only 27% of the individual plots where adults comprise 80 to 100% of cover. Together, one or the other life stage dominated in 67% of individual plots. These patterns were similar in 2007 (49% of plots with 0 to 20% garlic mustard cover due to adults, and 17% with 80 to

←

2008. Data were collected on seedlings and rosettes in the spring at 5 monitoring sites in 2005 (NE, WN, BP, FS, and PB) and at all 12 monitoring sites in 2006 to 2008. Sites are organized according to the rosette density patterns seen in the fall data. Two sites show strong cycling (one life stage is dominant each year) with rosette densities peaking every other year for all 4 yr. Four sites show cycling in the first 3 yr. WI has increasing population density and PB has decreasing population density as indicated by arrows. Two sites are relatively stable with little year to year variation in rosette population density (densities with SE overlap from year to year). Two sites show variation over time and don't clearly fit with any of the previous categories. BP = Baker Park, CR = Coon Rapids, CG = Cottage Grove, FS = Fort Snelling, HP = Hilloway Park, LL = Luce Line, NE = Nerstrand, PB = Pine Bend, PL = Plainview, WN = Warner Nature Center, WH = Westwood Hills, WI = Willmar.

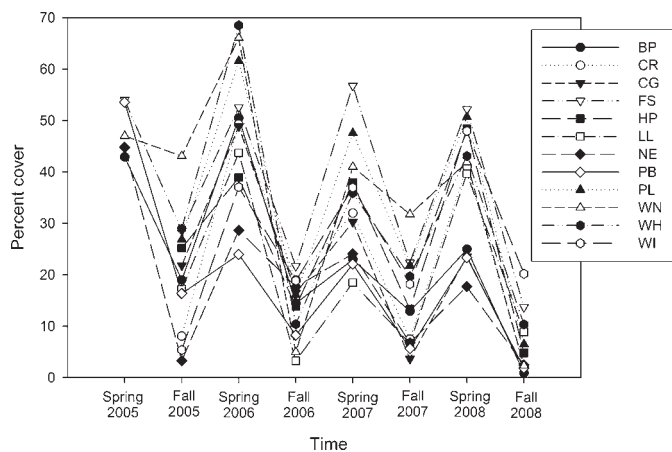


Figure 3. Mean visual percent cover of garlic mustard at each site over time. Spring garlic mustard cover is the total cover of adults and seedlings. Fall cover is cover of the rosettes. Data were collected in Minnesota in the spring at 5 monitoring sites in 2005 (BP, FS, NE, PB, and WN) and at all 12 monitoring sites in 2006 to 2008. BP=Baker Park, CR=Coon Rapids, CG=Cottage Grove, FS=Fort Snelling, HP=Hilloway Park, LL=Luce Line, NE=Nerstrand, PB=Pine Bend, PL=Plainview, WN=Warner Nature Center, WH=Westwood Hills, WI=Willmar.

100% cover due to adults, for a total of 66% of plots dominated by one life stage) and 2008 (37, 26, and 63%, respectively).

Our results indicate that at the individual plot level, garlic mustard populations tended to be dominated by one life stage or the other. Based on our data, half of the 12 sites monitored did not appear to display a cycle of dominance of one life stage over the other on average. However, even at these sites, several individual plots were cycling with either adult or seedling stages of garlic mustard. This phenomenon—if it occurs beyond Minnesota as well—lessens concerns, though still present, that critical life stage synchrony may not be available and may limit successful establishment of biological control insects. Knowledge of the life stage of a particular site, in addition to its importance to successful establishment of biological control agents, is important to determining the impacts of biological control agents post-release.

It will take several years of monitoring data to determine if a particular garlic mustard population is increasing, decreasing, or stable. Applying control methods can release remaining garlic mustard plants from intraspecific competition, and cause garlic mustard populations to no longer be governed by density-dependence and to exhibit complex and unpredictable changes in population density (Pardini et al. 2009). Monitoring garlic mustard populations over time after biological control insects are established will be especially important in determining whether these complex

population dynamics occur, or if there is a general decline in garlic mustard populations.

Fluctuations in Garlic Mustard Plant Height and Reproductive Output. Large natural fluctuations in annual garlic mustard reproductive output as determined by plant height and numbers of siliques were detected as height and silique production decreased from 2006 to 2007 with rebounds in 2008 (Figure 4a and 4b). The mean height of adult garlic mustard plants decreased at all 12 sites from 2006 to 2007 (Figure 4a). Mean heights ranged from 48 to 82 cm in 2006, but only 21 to 56 cm in 2007. Shorter garlic mustard plants then produced fewer siliques (Figure 4b). In 2006, silique production was high with 134 to 888 siliques m^{-2} , but by 2007 there were only 86 to 480 siliques m^{-2} (Figure 4b). The number of siliques produced per m^2 varied considerably from year to year depending on whether the silique-producing adult plants or the nonreproductive seedling stage was dominant. The mean number of siliques per garlic mustard stem ranged from 2 to 22 siliques $stem^{-1}$ (Figure 4c). Since plants were generally shorter in 2007 than 2006, they correspondingly produced fewer siliques per plant with 5 to 14 siliques $stem^{-1}$ in 2006 vs. 2 to 11 siliques $stem^{-1}$ in 2007 (Figure 4c). Environmental factors, such as below normal precipitation in 2007, were the likely cause of decreased growth and production of siliques at all the sites. From April to June 2007 there were only 13 cm of precipitation compared to the 30-yr (1971 to 2000) average April to June precipitation of 25 cm (Minnesota Climatology Working Group 2009). Other studies have found that June precipitation correlates positively with garlic mustard rosette density in October of that year and adult density in June of the next year (Hochstedler and Gorchov 2007; Slaughter et al. 2007).

To further characterize the population, adult stems were categorized as with or without siliques. When biological control is released, we expect that the insects will damage plants and cause a reduction in plant height and number of siliques (Gerber et al. 2007a,b). At most sites, more than 95% of the adult stems produced siliques (Figure 4d). With one exception, the sites had fewer than 10% barren stems. The abnormally high percentage of barren stems (26%) observed at HP in 2006 was due to early season *R. cathartica* control, which resulted in herbicide drift onto garlic mustard plants. This caused reduced and delayed silique development in many plants.

Relationship Between Garlic Mustard and Native Species. Our work supports previous findings that garlic mustard forms dense populations that negatively impact native species (Blossey et al. 2001; Nuzzo 1999; Stinson et al. 2007). There were consistent negative correlations between total garlic mustard cover in the spring (adults + seedlings) and native species richness in the spring (Pearsons correlation $P = 0.04, 0.27, 0.24$ and $R^2 =$

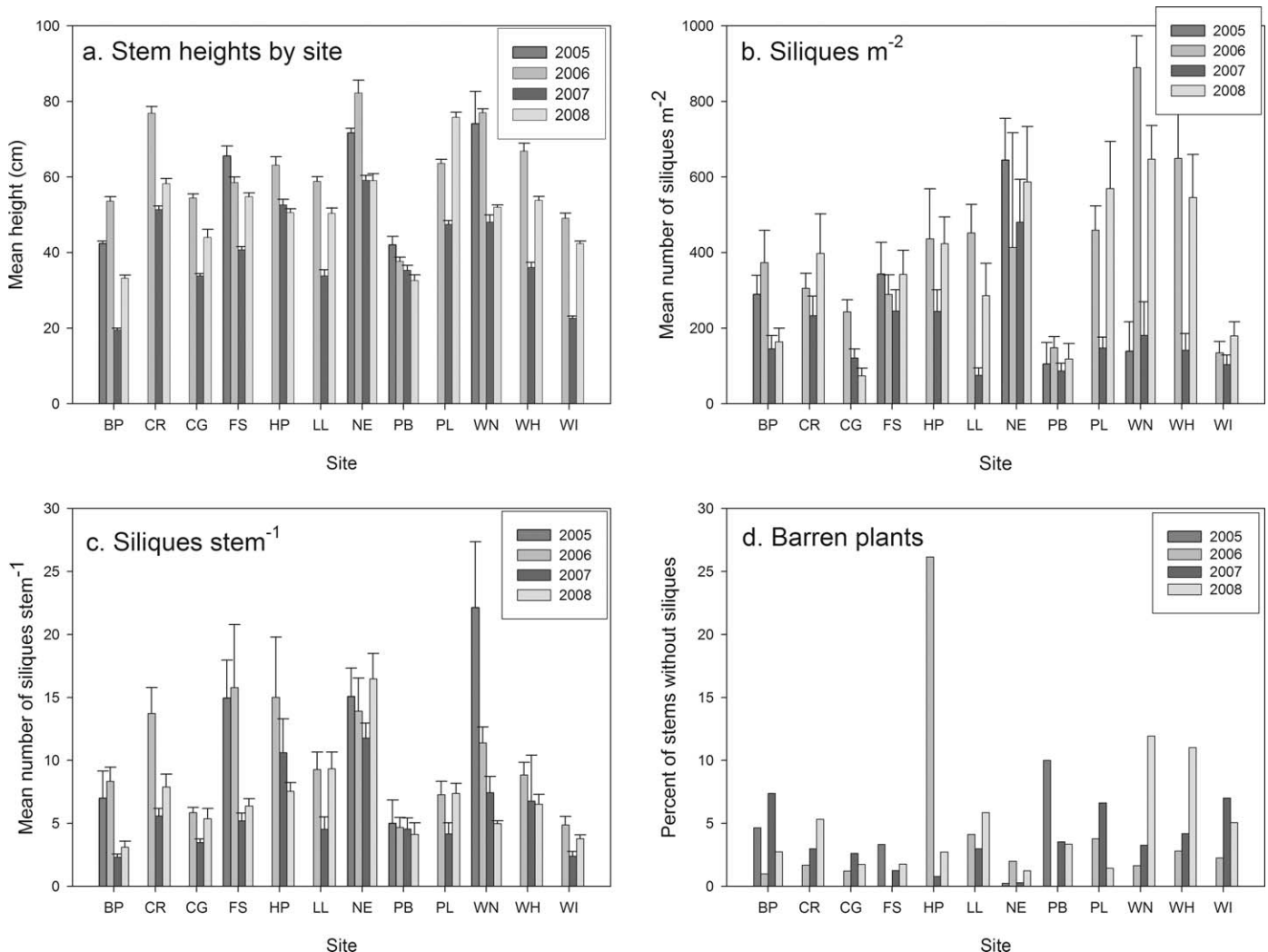


Figure 4. (a) Mean adult garlic mustard plant height (\pm standard error [SE]) by site, (b) mean number of siliques per m² (\pm SE) (an estimate of seed output at a site), (c) mean number of siliques per adult stem (\pm SE) (a measure of the fecundity of individual plants at a site), and (d) percent of adult garlic mustard stems without siliques present (the total number of sterile stems recorded at the site / total number of stems at the site \times 100%). Data were collected in Minnesota in the spring at 5 monitoring sites in 2005 (BP, FS, NE, PB, and WN) and at all 12 monitoring sites in 2006 to 2008. Note that the 2006 HP percent barren plants was high because many garlic mustard plants were impacted by herbicide drift from adjacent buckthorn treatments. BP = Baker Park, CR = Coon Rapids, CG = Cottage Grove, FS = Fort Snelling, HP = Hilloway Park, LL = Luce Line, NE = Nerstrand, PB = Pine Bend, PL = Plainview, WN = Warner Nature Center, WH = Westwood Hills, WI = Willmar.

−0.59, −0.34, and −0.36 for 2006, 2007, and 2008, respectively). There were also consistent negative correlations between total garlic mustard cover in the spring (adults + seedlings) and native species percent cover in the spring, although P values were nonsignificant (Pearsons correlation $P = 0.09, 0.28, 0.13$ and $R^2 = -0.51, -0.34, -0.47$ for 2006, 2007, and 2008, respectively). The highest species richness was found in the spring in these habitats. Sites differed strongly in disturbance history, from scientific and natural areas with late-successional, high diversity forests to highly disturbed, second-growth forests in

urban parks, which is likely a strong driver of native species cover and richness differences among the sites.

The most common native species found were *Galium aparine* L., *Circaea lutetiana* L., *Fraxinus pennsylvanica* seedlings, *Geum canadense* Jacq., *Ageratina altissima* (L.) King & H.E. Rob. var. *altissima*, *Pilea pumila* (L.) Gray, *Osmorhiza claytonia* (Michx.) C.B. Clarke, and *Laportea canadensis* (L.) Weddell. The most common nonnative species found were *R. cathartica*, *Glechoma hederacea* L., *Hackelia virginiana* (L.) I.M. Johnst., *Taraxacum officinale* G.H. Weber ex Wiggers, and *Solanum dulcamara* L. Spring

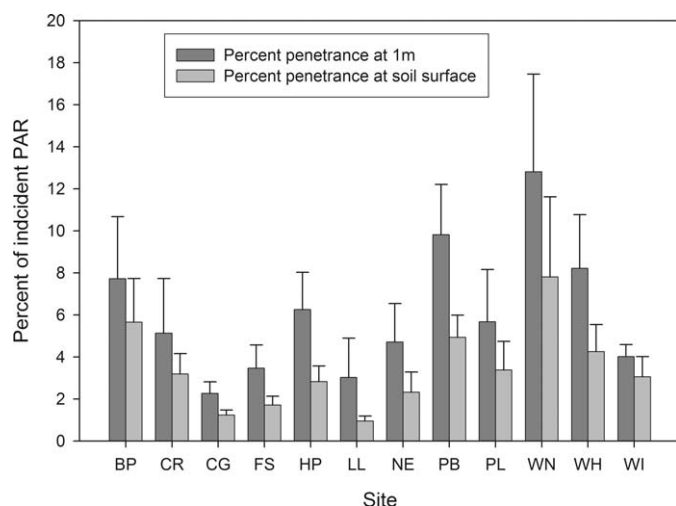


Figure 5. The mean percent of incident photosynthetically active radiation (PAR) penetrating to 1 m above the soil surface and at the soil surface (\pm standard error [SE]) as measured in August and September 2008 within 2 hr of solar noon at 12 sites in Minnesota. BP = Baker Park, CR = Coon Rapids, CG = Cottage Grove, FS = Fort Snelling, HP = Hilloway Park, LL = Luce Line, NE = Nerstrand, PB = Pine Bend, PL = Plainview, WN = Warner Nature Center, WH = Westwood Hills, WI = Willmar.

native species richness per 0.5-m² quadrat ranged from a low of 1.8 at BP in 2005 to a high of 7.1 at WI in 2008. Native species cover ranged from a low of 8.3% cover at Luce Line (LL) in 2008 to a high of 51.4% cover at WI in 2008. Nerstrand (NE) had the lowest nonnative species richness (the number of nonnative species present, not including garlic mustard) and cover with no nonnative species present in the spring from 2005 to 2007, and one nonnative species present in 2008 (*T. officinale*). The highest nonnative species richness per quadrat (1.8 nonnative species) and cover (27.8% cover) were found at CR in 2008 and were typically *G. hederacea* and *R. cathartica*.

The low numbers and cover of native species raise concerns for site restoration following garlic mustard control. If garlic mustard cover and population density are reduced, having a diverse and large native species population increases the chances that the site may move to a more native-species dominated state with minimal active restoration. However, once garlic mustard cover is high and native species richness is low for an extended period of time, it is much more likely that active restoration actions may be necessary to restore native species to the sites. Studies of herbicide treatments of garlic mustard have found that native species, such as spring ephemerals, increase in cover after garlic mustard is reduced (Carlson and Gorchov 2004), although the effect is not always consistent among years (Hochstedler et al. 2007).

Garlic Mustard, Leaf Litter, and Earthworms. Our results showed little evidence for earthworm impacts on garlic mustard populations through reduced litter layer (data not shown) in part due to the fact that the studies were not designed to directly study earthworm impacts. All sites were found to have significantly impacted litter layers, so comparisons with undamaged sites are lacking. It has been shown, however, that garlic mustard invasion can be facilitated by earthworm invasion (Blossey et al. 2005; Nuzzo et al. 2009). We used low depth of the litter layer and high cover of bare soil as indicators of earthworm disturbance. All sites had very low depth of litter with average depths of litter in the spring ranging from < 0.1 to 2.4 cm. These data suggest that earthworms may have been burying leaf litter at all 12 sites; however, more research would be needed to confirm this. Observed depths of litter were consistent with low depths of litter recorded by other researchers in Minnesota (depth of litter layer has been shown to decrease from 10 to 0 cm with the presence of earthworms (Hale et al. 2005)). There was no accumulated litter from previous years. The litter that was measured was recent leaf fall, indicative of earthworm activity. The low variation in depths of litter across sites made it difficult to detect any relationship between increased garlic mustard densities in sites with low depth of litter (data not shown, regression P values > 0.34 , $R^2 < 0.09$ in 2006, 2007, and 2008). The percent cover of bare ground did vary widely across sites, ranging from 0 to 84% of the ground cover in the spring. Even with a range of bare ground cover, there was no indication of increasing garlic mustard population density with increasing amounts of bare ground (data not shown, regression P values > 0.54 , $R^2 < 0.04$ in 2006, 2007, and 2008). The widespread occurrence of low levels of litter indicate that garlic mustard populations will continue to grow and spread, as low litter sites are ideal for garlic mustard seedling establishment (Bartuszevige et al. 2007).

Influence of Available PAR. We looked at the potential for light availability to explain site to site differences and the tendency for some sites to be dominated by one life stage in a given year. Measurements in August and September of 2008 showed all sites generally had low levels of percent of full sun PAR incident at the top of the garlic mustard canopy (1-m height, mean of $6 \pm 0.7\%$) or at the soil surface (mean of $3 \pm 0.4\%$). Percent incident PAR at either level did not differ among sites due to large variations within these low levels (one-way ANOVA at 1 m $P = 0.08$, $F_{11,228} = 1.69$; and soil surface $P = 0.07$, $F_{11,228} = 1.72$). Mean percent of full sun PAR incident at the 1-m level ranged from 2% at Cottage Grove (CG) to 13% at WN, and at ground level from 1% at LL and CG to 8% at WN (Figure 5). Each site generally had a few plots in an area of a canopy opening, with some openings

Table 2. Garlic mustard presence and types of insect feeding at 12 sites in Minnesota, 2005 to 2008. The percentage of plots with garlic mustard present out of the 20 plots at each of 12 study sites in Minnesota over 3 yr are presented (5 study sites in spring 2005, 12 study sites for all other dates). Of the plots with garlic mustard present, the percentages of those plots with various types of visual leaf damage estimates are listed by the type of feeding damage.

Time	Plots with garlic mustard present	Plots with feeding by this insect type (of plots with garlic mustard present)				Mean leaf removal
		Edge feeding	Holes	Leaf miner	Windowpane feeding	
<hr/>						
<div>%</div> <hr/>						
Spring 2005	100	96	98	31	4	1.6
Fall 2005	87	99	98	1	1	1.5
Spring 2006	98	96	97	31	9	1.5
Fall 2006	84	97	98	< 1	< 1	2.0
Spring 2007	99	100	100	33	0	1.8
Fall 2007	88	97	96	1	0	2.4
Spring 2008	99	100	98	12	4	2.3
Fall 2008	63	97	91	0	< 1	3.0

allowing 20 to 90% of ambient light to reach the plots, accounting for the large standard errors in the mean percentages of incident PAR.

The light measurements taken in August and September 2008 to characterize the sites were compared with the garlic mustard seedling and adult percent cover measures taken in June 2008 and rosette percent cover measures taken in October 2008. When the garlic mustard cover data for the 240 plots were regressed against the amount of incident PAR, there were no strong relationships. Regressions of garlic mustard cover vs. the amount of incident PAR at the soil surface showed no relationship with seedlings ($P = 0.26$, $R^2 = 0.005$) and rosettes ($P = 0.78$, $R^2 = 0.0003$), and a weak positive relationship with adults ($P = 0.003$, $R^2 = 0.032$). The pattern was similar for regressions vs. PAR at 1 m above the soil surface for seedlings ($P = 0.17$, $R^2 = 0.008$), adults ($P = 0.01$, $R^2 = 0.025$), and rosettes ($P = 0.41$, $R^2 = 0.003$). However, when mean garlic mustard cover and incident PAR levels were calculated for the 12 sites and the data analyzed at the site level, there was a significant relationship between light and seedling cover. The regression of the cover of garlic mustard seedlings against percent of PAR penetrating to the soil surface showed a negative relationship ($P = 0.01$, $R^2 = 0.49$), while the relationships with PAR and adult cover ($P = 0.12$, $R^2 = 0.22$, trending positive) and rosette cover ($P = 0.32$, $R^2 = 0.10$, trending negative) were nonsignificant. Regressions of garlic mustard cover against light at 1 m above soil surface were similar to those at the soil surface for seedlings ($P = 0.01$, $R^2 = 0.48$), adults ($P = 0.13$, $R^2 = 0.21$), and rosettes ($P = 0.25$, $R^2 = 0.13$). The seedling stage appeared to be most sensitive to the amount of available light. Adult garlic mustard plants showed greater percent cover in sites with higher available PAR, but cover of seedlings and rosettes showed the opposite pattern.

These PAR measurements were taken in August and September after the adults had died back. At sites with high adult cover, adults likely shaded out seedlings, causing the sites with high adult cover to have low cover of seedlings. The pattern of lower cover of seedlings likely persisted as the seedlings grew into rosettes.

Overall, sites were similar in the amount of light available, indicating that light alone is likely not driving site differences. Adult plants trended toward the expected pattern of increased growth with increased light (Meekins and McCarthy 2000; Myers et al. 2005). The negative relationship of seedlings and rosettes with light supports the density-dependent nature of garlic mustard (Pardini et al. 2009) with the adult plants shading out the younger plants and keeping the cover of younger plants low.

Garlic Mustard Herbivory Levels. While evidence of insect feeding was widespread, the actual amount of leaf damage was low (Table 2). Leaf feeding damage was found in 96 to 100% of plots in which garlic mustard was present. The most common forms of damage were edge or internal hole feeding present on over 91% of the plots that contained garlic mustard. Leaf mining and windowpane feeding also occurred, but at much lower levels. While edge and hole feeding was common in the spring and fall, leaf mining was a much less common occurrence in the fall, decreasing from 31% in the spring to only 1% in the fall, since most leaf mining was on adult plants. Across all sites, seasons, and years, the average amount of leaf area damaged due to insects was $2.0 \pm 0.03\%$. Across sites, mean leaf damage by insect feeding did not vary widely across years or seasons with a range of 1.5 to 3.0%. Among the individual sites, the lowest mean leaf removal was 1.0% at PB in spring 2006, while the highest was 5.5% at FS in fall 2008.

Notably, little insect damage was present on garlic mustard populations in Minnesota. The 2% leaf damage levels in Minnesota were similar to the 3% leaf damage levels reported in Michigan (Evans and Landis 2007). Releasing *C. scrobicollis* weevils for biological control would introduce a significant, new stressor to garlic mustard (Gerber et al. 2007b).

Implications for Biological Control. Garlic mustard plant population densities and dominant growth stage varied considerably from year to year, amplified in part by its biennial life cycle. At some sites, population fluctuations were due the cyclic changes in dominance between the seedling and adult stages. Understanding which cycle a site is in at the time of biological control agent introduction will improve chances for biological control insect establishment, which is most critical in the first releases when numbers of insects available for release will be limiting. Releases should be targeted matching the phenologies of the target plant, insect, and the method of insect release, e.g., releasing egg laying adults whose larvae need to burrow into stems or feed on seeds in a cycle where seedlings will dominate should be avoided. It is also important to take these population cycles into account when analyzing the impacts of management efforts. A decrease in adult plants from 1 yr to the next may simply be a result in this natural oscillation in life stage dominance.

Should biological control insects be released, we expect to see decreases in garlic mustard populations (Davis et al. 2006; Gerber et al. 2007a,b). If biological control insects are approved for release, insects will be released on 6 of the 12 monitoring sites while the other 6 will act as controls. This will allow us to separate changes in plant communities due to biological control insects from changes due to other biotic or abiotic factors. Individual species and functional groups have been found to vary in their responses to experimental removal of garlic mustard (McCarthy 1997; Stinson et al. 2007). Tree seedlings and native grasses are especially susceptible to the presence of garlic mustard and are some of the first species to increase after garlic mustard removal (McCarthy 1997; Stinson et al. 2007).

We found that garlic mustard in Minnesota is currently experiencing very little herbivory. This lack of insect stressors may be one reason why garlic mustard has been such a successful invader. We confirmed that garlic mustard is currently lacking significant insect or disease pressures, and has the resources available to complete its life cycle and produce abundant seed. It is anticipated that the introduction of biological control insects in the United States will stress the plants and result in smaller plants that produce fewer siliques, as reported by Gerber et al. (2007a,b), and the number of stems without siliques will increase. It will likely take several years of reduced garlic

mustard populations before impacts on the forest understory can be observed (Hochstedler et al. 2007).

After biological control release, the potential exists for large differences in native plant community recovery among the different sites. Some sites have high levels of disturbance (low litter levels, high nonnative species cover) while others have a more robust native plant community. For example, excluding garlic mustard, NE had no cover of other nonnative species measured in the spring for 3 yr. If garlic mustard decreases, there is a diverse native species community ready to expand. In contrast, BP had the lowest cover of native species and the highest diversity of nonnative species. Monitoring will indicate whether native species are able to reestablish in areas currently dominated by garlic mustard. Our studies encompassed a sufficiently robust range of disturbance levels, light levels, and other abiotic factors to enable us to adequately characterize potential impacts of the biological control agents should they be released, and determine whether those impacts are consistent across sites. Furthermore, this study demonstrates the wide variability in garlic mustard population dynamics, the potential for continued negative impacts of garlic mustard to the remaining native species, and the potential for restoration challenges should garlic mustard be reduced.

Sources of Materials

¹ LI-190SA point quantum light sensor and LI-1000 data logger, LI-COR Biosciences, 4647 Superior St., Lincoln, NE, 68504.

² 1-m LI-191SA line quantum light sensor and LI-189 visual display, LI-COR Biosciences, 4647 Superior St., Lincoln, NE, 68504.

Acknowledgments

Funding for this project was provided by the Minnesota Environment and Natural Resources Trust Fund as recommended by the Legislative-Citizen Commission on Minnesota Resources. Thank you to the land managers and owners of the 12 monitoring sites for their help and cooperation. G. Kasowski, M. Odima, B. Kinkaid, M. Carlson, M. Chandler, J. Carylton, C. Drake, A. Einck, K. Jung, N. Northrop, and A. Shackelford assisted with data collection in the field.

Literature Cited

- Bartuszevige, A. M., R. L. Hrenko, and D. L. Gorchov. 2007. Effects of leaf litter on establishment, growth and survival of invasive plant seedlings in a deciduous forest. *Am. Midl. Nat.* 158:472–477.
- Blossey, B. 1999. Before, during and after: the need for long-term monitoring in invasive plant species management. *Biol. Invasions* 1: 301–311.
- Blossey, B., V. A. Nuzzo, H. L. Hinz, and E. Gerber. 2001. Developing biological control of *Alliaria petiolata* (M. Bieb.) Cavara and Grande (garlic mustard). *Nat. Areas J.* 21:357–367.
- Blossey, B., V. A. Nuzzo, J. Maerz, and A. Dávalos. 2005. Ecosystem impacts of *Alliaria petiolata* (garlic mustard). Pages 1–3 *in*

- Proceedings: Symposium on the Biology, Ecology, and Management of Garlic Mustard (*Alliaria petiolata*) and European Buckthorn (*Rhamnus cathartica*). St. Paul, MN: USDA Forest Service.
- Bohlen, P. J., P. M. Groffman, T. J. Fahey, M. C. Fisk, E. Suarez, D. M. Pelletier, and R. T. Fahey. 2004. Ecosystem consequences of exotic earthworm invasion of north temperate forests. *Ecosystems* 7: 1–12.
- Carlson, A. M. and D. L. Gorchov. 2004. Effects of herbicide on the invasive biennial *Alliaria petiolata* (garlic mustard) and initial responses of native plants in a southwestern Ohio forest. *Restor. Ecol.* 12:559–567.
- Cavers, P. B., M. I. Heagy, and R. F. Kokron. 1979. The biology of Canadian weeds. 35. *Alliaria petiolata* (M. Bieb.) Cavara and Grande. *Can. J. Plant Sci.* 59:217–229.
- Davis, A. S., D. A. Landis, V. A. Nuzzo, B. Blossey, E. Gerber, and H. L. Hinz. 2006. Demographic models inform selection of biocontrol agents for garlic mustard (*Alliaria petiolata*). *Ecol. Appl.* 16: 2399–2410.
- Eschtruth, A. K. and J. J. Battles. 2009a. Acceleration of exotic plant invasion in a forested ecosystem by a generalist herbivore. *Conserv. Biol.* 23:388–399.
- Eschtruth, A. K. and J. J. Battles. 2009b. Assessing the relative importance of disturbance, herbivory, diversity, and propagule pressure in exotic plant invasion. *Ecol. Monogr.* 79:265–280.
- Evans, J. A. and D. A. Landis. 2007. Pre-release monitoring of *Alliaria petiolata* (garlic mustard) invasions and the impacts of extant natural enemies in southern Michigan forests. *Biol. Control* 42:300–307.
- Gerber, E., H. L. Hinz, and B. Blossey. 2007a. Interaction of specialist root and shoot herbivores of *Alliaria petiolata* and their impact on plant performance and reproduction. *Ecol. Entomol.* 32:357–365.
- Gerber, E., H. L. Hinz, and B. Blossey. 2007b. Impact of the belowground herbivore and potential biological control agent, *Ceutorhynchus scrobicollis*, on *Alliaria petiolata* performance. *Biol. Control* 42:355–364.
- Hale, C. M., L. E. Frelich, and P. B. Reich. 2005. Exotic European earthworm invasion dynamics in northern hardwood forests of Minnesota, USA. *Ecol. Appl.* 15:848–860.
- Hochstedler, W. W. and D. L. Gorchov. 2007. The effects of June precipitation on *Alliaria petiolata* (garlic mustard) growth, density and survival. *Ohio J. Sci.* 107:26–31.
- Hochstedler, W. W., B. S. Slaughter, D. L. Gorchov, L. P. Saunders, and H. H. Stevens. 2007. Forest floor plant community response to experimental control of the invasive biennial, *Alliaria petiolata* (garlic mustard). *J. Torrey Bot. Soc.* 134:155–165.
- Katovich, E. J. S., R. L. Becker, D. W. Ragsdale, and L. C. Skinner. 2005. Host range testing of garlic mustard (*Alliaria petiolata*) biocontrol insects in Minnesota. Pages 17–18 in *Proceedings: Symposium on the Biology, Ecology, and Management of Garlic Mustard (Alliaria petiolata) and European Buckthorn (Rhamnus cathartica)*. St. Paul, MN: USDA Forest Service.
- McCarthy, B. C. 1997. Response of a forest understory community to experimental removal of an invasive nonindigenous plant (*Alliaria petiolata*, Brassicaceae). Pages 117–130 in J. O. Luken and J. W. Thieret, eds. *Assessment and Management of Plant Invasions*. New York: Springer-Verlag.
- Meekins, J. F. and B. C. McCarthy. 2000. Responses of the biennial forest herb *Alliaria petiolata* to variation in population density, nutrient addition and light availability. *J. Ecol.* 88:447–463.
- Meekins, J. F. and B. C. McCarthy. 2002. Effect of population density on the demography of an invasive plant (*Alliaria petiolata*, Brassicaceae) population in a southeastern Ohio forest. *Am. Midl. Nat.* 147:256–278.
- Meekins, J. F., H. E. Ballard, and B. C. McCarthy. 2001. Genetic variation and molecular biogeography of a North American invasive plant species (*Alliaria petiolata*, Brassicaceae). *Int. J. Plant Sci.* 162: 161–169.
- Minnesota Climatology Working Group. 2009. <http://climate.umn.edu/text/historical/msppre.txt>. Accessed: March 4, 2009.
- Minnesota Department of Natural Resources. 2009. Species List for Minnesota. http://files.dnr.state.mn.us/ecological_services/plant_list9-25-02.pdf. Accessed: February 2, 2009.
- Myers, C. V., R. C. Anderson, and D. L. Byers. 2005. Influence of shading on the growth and leaf photosynthesis of the invasive non-indigenous plant garlic mustard [*Alliaria petiolata* (M. Bieb.) Cavara and Grande] grown under simulated late-winter to mid-spring conditions. *J. Torrey Bot. Soc.* 132:1–10.
- Nuzzo, V. A. 1999. Invasion pattern of the herb garlic mustard (*Alliaria petiolata*) in high quality forests. *Biol. Invasions* 1:169–179.
- Nuzzo, V. A., J. C. Maerz, and B. Blossey. 2009. Earthworm invasion as the driving force behind plant invasion and community change in northeastern North American forests. *Conserv. Biol.* 23:966–974.
- Oehlert, G. W. and C. Bingham. 2005. MacAnova 5.05. University of Minnesota. <http://www.stat.umn.edu/macanova>. Accessed: September 6, 2005.
- Pardini, E. A., J. M. Drake, J. M. Chase, and T. M. Knight. 2009. Complex population dynamics and control of the invasive biennial *Alliaria petiolata* (garlic mustard). *Ecol. Appl.* 19:387–397.
- Prati, D. and O. Bossdorf. 2004. Allelopathic inhibition of germination by *Alliaria petiolata* (Brassicaceae). *Am. J. Bot.* 91:285–288.
- Rodgers, V. L., K. A. Stinson, and A. C. Finzi. 2008. Ready or not, garlic mustard is moving in: *Alliaria petiolata* as a member of eastern North American forests. *BioScience* 58:426–436.
- Slaughter, B. S., W. W. Hochstedler, D. L. Gorchov, and A. M. Carlson. 2007. Response of *Alliaria petiolata* (garlic mustard) to five years of fall herbicide application in a southern Ohio deciduous forest. *J. Torrey Bot. Soc.* 134:18–26.
- Statistix 7. 2000. Statistix version 7.0. Tallahassee, FL: Analytical Software.
- Stinson, K. A., S. A. Campbell, J. R. Powell, B. E. Wolfe, R. M. Callaway, G. C. Thelen, S. G. Hallett, D. Prati, and J. N. Klironomos. 2006. Invasive plant suppresses the growth of native tree seedlings by disrupting belowground mutualisms. *PLoS Biol* 4: e140.
- Stinson, K., S. Kaufman, L. Durbin, and F. Lowenstein. 2007. Impacts of garlic mustard invasion on a forest understory community. *Northeast. Nat.* 14:73–88.
- Winterer, J., M. C. Walsh, M. Poddar, J. W. Brennan, and S. M. Primak. 2005. Spatial and temporal segregation of juvenile and mature garlic mustard plants (*Alliaria petiolata*) in a central Pennsylvania woodland. *Am. Midl. Nat.* 153:209–216.
- Wolfe, B. E., V. L. Rodgers, K. A. Stinson, and A. Pringle. 2008. The invasive plant *Alliaria petiolata* (garlic mustard) inhibits ectomycorrhizal fungi in its introduced range. *J. Ecol.* 96:777–783.

Received July 27, 2009, and approved November 3, 2009.