UNIVERSITY OF CINCINNATI

May 20 , 20 08

I, Cory C. Christopher **,**

hereby submit this as part of the requirements for the degree of:

Doctor of Philosophy

in:

Biological Sciences

It is entitled:

"Effects of invasive Amur honeysuckle (Lonicera maackii) and white-tailed deer (Odocoileus virginianus) on native plants, leaf litter communities, and soil"

Approved by:

Guy N. Cameron

Theresa M. Culley

Eric F. Maurer

David L. Gorchov

George W. Uetz

Effects of invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) on native plants, leaf litter communities, and soil

A dissertation submitted to the

Division of Research and Advanced Studies
of the University of Cincinnati

in partial fulfillment of the
requirements for the degree of

DOCTORATE OF PHILOSOPHY (Ph.D.)

in the Department of Biological Sciences
of the College of Arts & Sciences

2008

by

Cory C. Christopher

B.S., University of Georgia, 2000
M.S., University of Georgia, 2002

Committee Chair: Guy N. Cameron

Abstract

In the Midwest US, invasion by Amur honeysuckle (*Lonicera maackii*) reduces diversity, growth, and reproduction of native plants, and browsing by overabundant white-tailed deer (*Odocoileus virginianus*) may compound these impacts. Few studies, however, have determined whether these species act in concert to alter native plants, or whether these species impact litter invertebrate communities or forest soil. Using a combination of exclusion of white-tailed deer and removal of Amur honeysuckle, I measured individual and combined impacts of these species on diversity, abundance, and community composition of understory herbs and litter-dwelling invertebrates. I also examined whether deer or honeysuckle affected litter substrate composition, litter depth, soil compaction, and soil microbial activity. Amur honeysuckle, but not white-tailed deer, altered composition of forest understory herb and invertebrate communities, and had variable, but significant, effects on abundance and diversity of different herb species and invertebrate orders. Deer reduced invertebrate abundance but did not affect diversity or composition of invertebrates. Neither deer nor honeysuckle affected composition of litter substrate, mass of leaf fall, or litter depth. Leaf decomposition was similar across treatments. Soil compaction was greater in plots containing either deer or honeysuckle, but removal of honeysuckle and exclusion of deer reduced this effect. There were no interactions between deer and honeysuckle on decomposition of leaf litter or compaction of soil. Microbial activity was greater in homogenized topsoil when topped with decomposing leaves of honeysuckle than when under leaves of sugar maple. However, microbial activity in soil taken from an invaded area of forest was similar to that from adjacent uninvaded areas. The variable effects of white-tailed deer and Amur honeysuckle on different taxa and levels of organization requires that management of these species utilize habitat and taxa-specific control and restoration strategies. Future studies

addressing impacts of invasive plants or over-abundant ungulates would benefit by combining population-level questions with higher-level questions asking how these pest species alter community structure or ecosystem functions.

Acknowledgements

I first want to thank Guy Cameron, Theresa Culley, Eric Maurer, David Gorchov, and George Uetz for serving on my PhD advisory committee, and for providing invaluable assistance in my experimental design, implementation, and interpretation of my study. These thanks go especially to Guy Cameron, my PhD advisor and committee chair who always allowed me to barge into his office with random questions and complaints. I also must thank Stephen Matter for offering helpful advice on statistical analysis and data interpretation. I could not have identified many of the more difficult plant species without Majie Becus and Denis Conover, and Jenai Rutledge helped me greatly with spider identification. I also appreciate the assistance of Brian Kinkle and Jodi Shann with soil methodology and data interpretation. I owe much to the many field and lab assistants who assisted me in all aspects of my research, including Jessica Brzyski, Devan Capella, Ali Cooperman, Michael Cramer, Elizabeth Dame, Greg Klein, Jackie Kleinhans, Jessica Lawrence, Omar Mirza, Scott Puthoff, and David Wright. Outside of school, my parents offered much emotional support and have always encouraged me to cut my own path. I must also extend many thanks to my in-laws who supplied enumerable meals, countless hours of entertainment, and endless emotional support. Finally, I want to thank my wife Liz for offering keen scientific advice and for always reminding me to keep my head up so that I wouldn't miss the bigger picture.

Table of Contents

List of figures

Chapter 2

Chapter 3

Chapter 4

List of tables

Chapter 2

Chapter 3

Chapter 4

Chapter one: Introduction

In the US, invasion of exotic plants into new, previously unoccupied habitat costs agriculture, forestry, and public health industries billions of dollars per year (Pimentel et al. 2005). With these economic losses, exotic plant invasion has resulted in a suite of negative impacts on ecosystem function (Mack et al. 2000). For example, once introduced into a new habitat, invasive plants can alter rates of primary productivity, interfere with nutrient cycling, hybridize with native species, and outcompete native plants for water, nutrients, and light (Ramakrishnan and Vitousek 1989, Levin et al. 1996). These effects are causing a major problem for land managers, and increased worldwide travel, urbanization, and agriculture have increased the spread of exotic species over the entire globe (Di Castri 1989).

One such invasive species is Amur honeysuckle (*Lonicera maackii*), a shrub originally introduced to North America from northeastern Asia in 1896 for use in horticulture. In the 112 years since its introduction, it has invaded more than 24 states and at least 34 counties in Ohio (Trisel 1997). Where established, Amur honeysuckle reduces abundance (Hutchinson and Vankat 1997), richness (Collier et al. 2002), fecundity and fitness (Gould and Gorchov 2000), and growth and seed production (Miller and Gorchov 2004) of native herbs. It also reduces density and species richness (Collier et al. 2002), and survival (Gorchov and Trisel 2003) of tree seedlings, and its shallow, extensive root system may reduce water and nutrient availability to native plants (Hutchinson and Vankat 1997). Because it forms a dense canopy over the forest floor, Amur honeysuckle also reduces light penetration (Hutchinson and Vankat 1997) and may reduce complexity of the litter layer (Buddle et al. 2004).

These impacts to native plant communities and structure of leaf litter also may affect communities of litter-dwelling invertebrates. For example, diversity and abundance of litter

invertebrates are sensitive to changes in litter quality and microclimate (Badejo et al. 1998), litter complexity (Bultman and Uetz 1984), and litter depth (Antvogel and Bonn 2001). In addition, litter-dwelling invertebrates also are influenced by changes in plant diversity (Strong et al. 1984), structure (Gibson et al. 1992) and height of plants (Kruess and Tscharntke 2002). Understanding how changes in plant and litter communities affect litter invertebrates is important because invertebrates play essential roles in ecosystem processes such as litter decomposition and nutrient cycling (Seastedt and Crossley 1984, Kremen et al. 1993, Kim 1993, Hunter et al. 2003).

Few studies have sought to identify how effects of Amur honeysuckle on native plant and invertebrate communities could lead to changes in the structure of soil microbial communities, and whether these impacts translate to changes in ecosystem functions such as litter decomposition (Hector et al. 2000). For example, loss of plant diversity could reduce rates of litter decomposition due to increased C:N ratios of litter (Hector et al. 2000). Alternatively, rates of litter decomposition and nutrient cycling can also be affected by changes in the structure of communities of litter invertebrates (Bradford et al. 2002, Hunter et al. 2003) or soil fauna (Coûteaux et al. 1995, Kourtev et al. 2002) caused by invasion of exotic plants. Direct effects such as allelopathy may also influence the success of invasive plants (Hierro and Callaway 2003). For instance, allelopathic compounds in leaves and roots of Amur honeysuckle (Trisel 1997, Dorning and Cipollini 2006, Cipollini et al. 2008) could alter microbial activity in soil, and this could impact rates of litter decomposition. Altered rates of litter decomposition and nutrient cycling, in turn, lead to further alteration of plant species composition or diversity (Hooper and Vitousek 1997, Ehrenfeld and Scott 2001).

Management of Amur honeysuckle usually involves cutting the shrubs at the base, applying an herbicide such as glyphosate, or both (Conover and Geiger 1993). Removal of Amur

honeysuckle increased fitness of native herbs (Gould and Gorchov 2000) and survival of native tree seedlings (Hartman and McCarthy 2004), but without protection from herbivory from white-tailed deer (*Odocoileus virginianus*), removal of Amur honeysuckle reduced growth of seedlings of white oak (*Quercus alba*) and sugar maple (*Acer saccharum*; Gorchov and Trisel 2003). Whether white-tailed deer interact with Amur honeysuckle to affect plant, invertebrate, and soil communities is unknown, but understanding this relationship could provide information to land managers who maintain areas impacted by both Amur honeysuckle and white-tailed deer.

The range of white-tailed deer extends across all of the US, but white-tailed deer are absent or rare in Utah, Nevada, and California (Smith 1991). In the eastern portion of their range, deer entirely overlap the range of Amur honeysuckle. As generalist feeders, populations of white-tailed deer increase with forest fragmentation (Smith 1991), especially in areas where agriculture provides particularly high-quality habitat (Torgerson and Porath 1984). These areas are prevalent across the Midwest, where density of deer has been rapidly increasing since the 1920's (Iverson and Iverson 1999). Especially where they occur in high densities, white-tailed deer have a wide array of direct impacts on communities of native herbaceous and woody plants (Russell et al. 2001).

Browsing by overabundant white-tailed deer reduces number of overstory stems and seedlings of native trees (Healey 1997), decreases plant growth, reproduction, and density (Rooney 1997), and may lower diversity of native herbaceous vegetation (Rooney and Waller 2003). Impacts to plant communities are reduced at densities < 7 deer/km^2 (Augustine et al. 1998), and impacts to individual plants are reduced at 5 – 10 deer/km^2 (Augustine and Frelich 1998), but densities of white-tailed deer have exceeded 120/km^2 in some parks in southwestern Ohio (Conover 2007). In heavily grazed areas, effects on herb communities can last more than

30 years after deer populations have been reduced (Balgooyen and Waller 1995). Grazing by over-abundant white-tailed deer results in browse-tolerant plants whose litter contains high ratios of C:N and is resistant to decomposition and mineralization (Augustine and McNaughton 1998), potentially increasing depth and complexity of leaf litter. By reducing plant abundance and diversity and altering structure of the leaf litter, white-tailed deer also can indirectly affect native invertebrate and soil communities (Hector et al. 2000, Rooney and Waller 2003).

Despite similar impacts of Amur honeysuckle and white-tailed deer on native plant, invertebrate, and soil communities, few studies have examined their combined effects on these systems. Gorchov and Trisel (2003) demonstrated that biomass and stem length of sugar maple (*Acer saccharum*) seedlings increased after removal of Amur honeysuckle shoots and protection from white-tailed deer. When unprotected from white-tailed deer, however, removal of Amur honeysuckle shoots reduced total biomass, root:shoot ratio, and leaf area of sugar maple seedlings. Further, biomass, stem length, and leaf area of *Q. rubra* seedlings were greater when roots of Amur honeysuckle were severed by trenching and above-ground shoots of Amur honeysuckle remained. These responses were attributed to protection of seedlings by Amur honeysuckle from grazing by white-tailed deer (Gorchov and Trisel 2003). For land managers, these data suggest that successful restoration of *Acer saccharum* and *Quercus rubra* after removal of Amur honeysuckle will also require management of white-tailed deer.

Hartman and McCarthy (2004), however, found a negative effect of Amur honeysuckle on survival of *Q. muehlenbergii* (Chinkapin oak), *Juglans nigra* (black walnut), *Prunus serotina* (black cherry), *Fraxinus pennsylvanica* (green ash), *Cornus floridana* (flowering dogwood), and *Cerci canadensis* (red bud) seedlings. In their study, survival of seedlings was not enhanced by

protection from herbivory by white-tailed deer (Hartman and McCarthy 2004), suggesting that deer exclusion was not necessary.

In the current study, I examined the individual and interactive effects of white-tailed deer and Amur honeysuckle on species composition, diversity, and richness of forest floor herbs, and assessed their impact on reproduction and abundance of individual herb species and different herb growth forms (Chapter 2). Because of the sensitivity of litter invertebrates to changes in the plant community (Murdoch et al. 1972, Strong et al. 1984), I also examined whether white-tailed deer and Amur honeysuckle, independently and together, affected diversity, abundance, and community composition of litter-dwelling invertebrates (Chapter 3). I evaluated whether depth and composition of the litter layer Ire impacted by presence of deer and/or honeysuckle, and whether such changes affected diversity or abundance of litter-dwelling invertebrates. Further, I tested whether activity of soil microbes differed under decomposing leaves of Amur honeysuckle compared to leaves of *A. saccharum* in pots in a controlled greenhouse. I compared these results to microbial activity in soil from forest stands invaded by Amur honeysuckle, stands from which Amur honeysuckle had been removed and stands which had never been invaded. Lastly, I examined whether Amur honeysuckle and/or white-tailed deer altered penetrability of forest soil or decomposition of native leaf litter, and whether exclusion of white-tailed deer and removal of Amur honeysuckle alleviated these effects (Chapter 4).

Literature Cited

Antvogel, H. and A. Bonn. 2001. Environmental parameters an microspatial distribution of insects: a case study of carabids in an alluvial forest. Ecography 24:470-482.

Augustine and Frelich 1998 Effects of white-tailed deer on populations of an understory forb in fragmented deciduous forests. Conservation Biology 12:995-1004.

Augustine, D.J. and S.J. McNaughton. 1998. Ungulate effects on the functional species composition of plant communities: herbivore selectivity and plant tolerance. Journal of Wildlife Management 62:1165-1183.

Badejo, M.A., T.I. Nathaniel, and G. Tian. 1998. Abundance of springtails (Collembola) under four agroforestry tree species with contrasting litter quality. Biology and Fertility of Soils 27:15-20.

Balgooyen, C.P. and D.M. Waller. 1995. The use of *Clintonia borealis* and other indicators to gauge impacts of white-tailed deer on plant communities in northern Wisconsin, USA. Natural Areas Journal. 15:308-318.

Bradford, M.A., G.M. Tordoff, T. Eggers, T. Hefin Jones, and J.E. Newington. 2002. Microbiota, fauna, and mesh size interactions in litter decomposition. Oikos 99:317-323.

Buddle, C.M., S. Higgins, and A.L. Rypstra. 2004. Ground-dwelling spider assemblages inhabiting riparian forests and hedgerows in a agricultural landscape. The American Midland Naturalist 151:15-26.

Bultman, T.L. and G.W. Uetz. 1984. Effect of structure and nutritional quality of litter on abundances of litter-dwelling arthropods. The American Midland Naturalist 111:165-172.

Cipollini, D., R. Stevenson, S. Enright, A. Eyles, and P. Bonello. 2008. Phenolic metabolites in leaves of the invasive shrub, *Lonicera maackii*, and their potential phytotoxic and anti-herbivory effects. Journal of Chemical Ecology 34:144-152.

Collier, M.H. and J.L. Vankat. 2002. Diminished plant richness and abundance below *Lonicera maackii*, and invasive shrub. The American Midland Naturalist 147:60-71.

Conover, D.G. and D. R. Geiger. 1993. Glycophosphate controls *L. maackii* in native woodland restoration (Ohio). Restoration and Management Notes 11:2.

Conover, D.G. 2007. Impact of deer on vegetation in Miami Whitewater Forest, Mitchell Memorial Forest and Shawnee Lookout Park. Contractual Report, Hamilton Country Parks, Ohio.

Coûteaux, M.M., P. Bottner, and B. Berg. 1995. Litter decomposition, climate and litter quality. Trends in Ecology and Evolution 10:63-66.

di Castri, F. 1989. History of biological invasions with special emphasis on the Old World. *In*: Drake, J.A., H.A. Mooney, F. di Castri, R.H. Groves, F.J. Kruger, M. Rejmanek, and M. Williamson (*Eds.*) Scope 37: Biological Invasions: a Global Perspective. John Wiley & Sons Ltd. New York.

Dorning, M. and D. Cipollini. 2006. Leaf and root extracts of the invasive shrub, *Lonicera maackii*, inhibit seed germination of three herbs with no autotoxic effects. Plant Ecology 184:287-296.

Ehrenfeld, J.G. and N. Scott. 2001. Invasive species and the soil: Effects on organisms and ecosystem processes. Ecological Applications 11:1259-1260.

Gibson, C.W.D., V.K. Brown, L. Losito, and G.C. McGavin. 1992. The response of invertebrate assemblies to grazing. Ecography 15:166-176.

Gorchov, D.L. and D.E. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology 166: 13-24.

Gould, A.M.A. and D.L. Gorchov. 2000. Effects of the exotic invasive shrub *Lonicera maackii*, *L. maackii*, on survival and fecundity of native forest annual herbs. The American Midland Naturalist 144:36-50.

Hartman, K.M. and B.C. McCarthy. 2004. Restoration of a forest understory after the removal of an invasive shrub, *L. maackii* (*Lonicera maackii*). Restoration Ecology 12:154-165.

Healey, W.M. 1997. Influence of deer on structure and composition of oak forests in central Massachusetts. pp. 249-266 *In* McShea, W.J., H.B. Underwood, and J.H. Rappole (Eds.). The Science of Overabundance: Deer Ecology and Population Management. Smithsonian Institution Press. Washington D.C.

Hector, A., A.J. Beale, A. Minns, S.J. Otway, and J.H. Lawton. 2000. Consequences of the reduction of plant diversity for litter decomposition: effects through litter quality and microenvironment. Oikos 90: 357-371.

Hierro, J.L. and R.M. Callaway. 2003. Allelopathy and exotic plant invasion. Plant and Soil 256:29-39.

Hooper, D.U. and P.M. Vitousek. 1997. The effects of plant composition and diversity on ecosystem processes. Science 277:1302-1305.

Hunter, M.D., S. Adl, C.M. Pringle, and D.C. Coleman. 2003. Relative effects of macroinvertebrates and habitat on the chemistry of litter during decomposition. Pedobiologia 47:101-115.

Hutchinson, T.F. and J.L. Vankat. 1997. Invasibility and effects of *L. maackii* in southwestern Ohio forests. Conservation Biology 11: 1117-1124.

Iverson, A.L. and L.R. Iverson. 1999. Spatial and temporal trends of deer harvest and deer-vehicle accidents in Ohio. Ohio Journal of Science 99:84-94.

Kim, K.C. 1993. Biodiversity, conservation and inventory: why insect matter. Biodiversity and Conservation 2:191-214.

Kourtev, P.S., J.G. Ehrenfeld, and M. Häggblom. 2002. Exotic plant species alter the microbial community structure and unction in the soil. Ecology 83:3152-3166.

Kremen, C., R.K. Colwell, T.L. Erwin, D.D. Murphy, R.F.Noss, and M.A. Sanjayan. 1993. Terrestrial arthropod assemblages: their use in conservation planning. Conservation Biology 7:796-808.

Kruess, A. and T. Tscharntke. 2002. Contrasting responses of plant and insect diversity to variation in grazing intensity. Biological Conservation 106:293-302.

Levin, D.A., J. Francisco-Ortega, and R.K. Jansen. 1996. Hybridization and the extinction of rare plant species. Conservation Biology 10:10-16.

Mack, R.N., D. Simberloff, W.M. Lonsdale, H. Evans, M. Clout, and F.A. Bazzaz. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. Ecological Applications 10: 689-710.

Miller, K.E. and D.L. Gorchov. 2004. The invasive shrub, *Lonicera maackii*, reduces growth and fecundity of perennial forest herbs. Oecolgia 139:359-375.

Murdoch, W.W., F.C. Evans, C.H. Peterson. 1972. Diversity and pattern in plants and insects. Ecology 53:819-829.

Pimentel, D., R. Zuniga, and D. Morrison. 2005. Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecological Economics 52:273-288.

Ramakrishnan, P.S. and P.M. Vitousek. 1989. Ecosystem-level processes and the consequences of biological invasions. *In*: Drake, J.A., H.A. Mooney, F. di Castri, R.H. Groves, F.J. Kruger, M. Rejmanek, and M. Williamson (*Eds.*) Scope 37: Biological Invasions: a Global Perspective. John Wiley & Sons Ltd. New York.

Rooney, T.P. 1997. Escaping herbivory: refuge effects on the morphology and shoot demography of the clonal forest herb *Maianthemum canadense*. Journal of the Torrey Botanical Society 124:280-285.

Rooney, T.P., and D.M. Waller. 2003. Direct and indirect effects of white-tailed deer in forest ecosystems. Forest Ecology and Management 181:165-176.

Russell, F.L., D.B. Zippin, and N.L. Fowler. 2001. Effects of white-tailed deer (*Odocoileus virginianus*) on plants, plant populations and communities: a review. The American Midland Naturalist 146:1-26.

Seastedt, T.R. and D.A. Crossley, Jr. 1984. The influence of arthropods on ecosystems. BioScience 34:157-161.

Smith, W.P. 1991. *Odocoileus virginianus*. The American Society of Mammalogists, Mammalian Species no.388:1-13.

Strong, D.R., J.H. Lawton, and R. Southwood. 1984. Insects on Plants, Community Patterns and Mechanisms. Harvard University Press. Cambridge, Massachusetts.

Torgerson, O. and W.R. Porath. 1984. White-tailed populations and habitats of the Midwest oak/hickory forests. Pp.411-426 i*n*: L.K. Halls (*Ed.*) White-Tailed Deer: Ecology and Management. Stackpole Books. Pennsylvania.

Trisel, D.E. 1997. The invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae): factors contributing to its success and its effect on native species. Ph.D. Dissertation, Miami University, Oxford, Ohio.

Waller, D.M. and W.S. Alverson. 1997. The white-tailed deer: a keystone herbivore. Wildlife Society Bulletin 25:217-226.

Chapter Two

Effects of Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) on community composition, abundance, and growth of forest herbs

Abstract: In the Midwest US, invasion by *Lonicera maackii* (Amur honeysuckle) has reduced diversity, growth, and reproduction of native plants, effects that may be further compounded by browsing by overabundant white-tailed deer (*Odocoileus virginianus*). Although most studies have focused on these two species independently, few have determined whether they act in concert to alter the plants in their shared community, despite their overlapping ranges across the eastern US and southeastern Canada. Using a combination of exclusion of *O. virginianus* and removal of *L. maackii*, we measured the combined impacts of these species on diversity and species composition of an understory herb community in southwestern Ohio. We determined whether effects from *O. virginianus* and *L. maackii* were interactive, were similar at different levels of organization, and whether removal of *O. virginianus* and/or *L. maackii* allowed recovery of impacted areas. We found that *O. virginianus* and *L maackii* altered composition of forest understory herb communities, and had variable, but significant, effects on abundance of different species and growth forms of herbs. We also found that both *L. maackii* and *O. virginianus* reduced leaf number and stem height of *Maianthemum racemosa* (false Solomon's seal), and a significant *L. maackii* x *O. virginianus* interaction revealed that leaf number and stem height were not reduced by *O. virginianus* when *L. maackii* was present. These data demonstrate the importance of integrating different levels of organization when measuring impacts of invasive plants or overabundant herbivores, as effects visible at one level were not necessarily obvious at another. Additionally, effects attributable to *L. maackii* may have masked

those caused by *O. virginianus*. For restoration of native forest communities impacted by invasive species and overabundant herbivores, particularly ungulates, measurement of how much of the total community impact is due to each species individually, and how much may be due to their interactions is recommended.

Key Words: *Odocoileus virginianus*, *Lonicera maackii*, interactions, white-tailed deer, Amur honeysuckle, invasive species, herbivory, plant diversity, community composition

Introduction

Invasive plants impart a suite of impacts on communities and ecosystems that they invade (Vitousek 1990) and they create major economic and environmental problems for land managers (Hobbs and Humphries 1995, Pimental et al. 2005). If these impacts are to be controlled and if successful management is to be implemented, it is important to understand the specific structural and functional effects that invasive plants have on native ecosystems (Hartman and McCarthy 2004, Yates et al. 2004). Such understanding also includes determining whether native species compound effects of invasives (Zavaleta et al. 2001, White et al. 2006). While it may not always be wise to delay management until all ecological effects of an invasive species are understood (Simberloff 2003), it also is important to not implement costly control efforts prematurely that could ultimately fail because of unknown or unanticipated effects from interactions with native species (White et al. 2006).

Herbivores, for example, interact in various ways with invasive plants (Maron and Vilà 2001). Herbivores may increase the spread of invasive plants by reducing their native competitors (Edwards et al. 2000, Kellogg and Bridgham 2004) or may limit their spread by grazing on them (i.e., biotic resistance, Creed and Sheldon 1995, Vilà and D'Antonio 1998, Case and Crawley 2000). While numerous studies have noted impacts of invasive plants or herbivores

separately (D'Antonio and Meyerson 2002, Huntley 1991), community effects due to interactions between invasive plants and native herbivores are not well understood, and may not be simply additive. For example, invasive wheat grass (*Pseudoroegeneria spicata*) and introduced cattle, goats, and rodents in Hawaii each reduced growth and survival of seedlings of the native lama tree (*Diospyros sandwicensis*; Cabin et al. 2000). However, *P. spictata* also increased survival of native seedlings if protected from cattle and goats, perhaps because shading increased concentration of leaf nitrogen in *D. sandwicensis* seedlings (Cabin et al. 2000).

In the eastern US, Amur honeysuckle (*Lonicera maackii*), an invasive shrub introduced to North America from Asia for horticulture in 1896 (Luken and Thieret 1995), has invaded more than 24 eastern states and has become naturalized in at least 34 counties in Ohio (Trisel 1997). *Lonicera maackii* reduces abundance (Hutchinson and Vankat 1997), diversity (Collier et al. 2002), fecundity and fitness (Gould and Gorchov 2000), and growth and seed production (Miller and Gorchov 2004) of native herbs in eastern deciduous forests. In addition, *L. maackii* reduces density and species richness (Hutchinson and Vankat 1997, Collier et al. 2002), and survival (Gorchov and Trisel 2003, Hartman and McCarthy 2004) of tree seedlings, as well as the rate of radial and basal area growth of overstory trees (Hartman and McCarthy 2007). Because *L. maackii* leafs out earlier in spring than native trees and shrubs (Trisel 1997), shade intolerant herbs such as spring perennials that grow and reproduce in early spring before the tree canopy develops may be particularly affected by *L. maackii* (Gould and Gorchov 2000), although this is not always the case (Miller and Gorchov 2004). Management of *L. maackii* typically includes removal of shrub crowns, application of herbicides such as glyphosate, or a combination of these methods (Conover and Geiger 1993, Trisel 1997, Hartman and McCarthy 2004).

Lonicera maackii is restricted to eastern North America where its range is overlapped by white-tailed deer (*Odocoileus virginianus*), the dominant large herbivore in this area (Myers et al. 2004). *Odocoileus virginianus* is a habitat generalist whose populations typically increase with forest fragmentation (Smith 1991). Agricultural areas that provide particularly high-quality habitat (Torgerson and Porath 1984) are prevalent across the Midwestern Agricultural Region, including Ohio, where density of deer has rapidly increased since the 1920's (Iverson and Iverson 1999). In some metro parks in southwestern Ohio, populations exceed 120 / km^2 and damage to vegetation is substantial (Conover 2007). Effects of *O. virginianus* on native vegetation differ depending on their population density. For example, negative impacts on herb populations due to grazing by deer are reduced at densities <7 deer/km^2 (Augustine et al. 1998), but impacts on individual plant growth are reduced at 5 – 10 deer/km^2 (Augustine and Frelich 1998). High density of *O. virginianus* reduces number of overstory stems and seedlings (Healey 1997), as well as size of herbaceous leaves and number of flowering shoots (Rooney 1997). Such effects upon native vegetation pose serious threats to restoration of endangered or threatened species (Peck and Stahl 1997).

Effects of herbivory by *O. virginianus* on forest vegetation vary seasonally, particularly in agricultural regions, since they move from forests to forage in fields and agricultural areas during summer (Smith 1991, Strole and Anderson 1992). Because of seasonal differences in foraging activity of *O. virginianus*, impacts on spring perennials may be more pronounced than impacts on summer perennials. Impacts also are species-specific, with *O. virginianus* preferring some species, particularly those in the families Liliaceae and Ranunculaceae (Frankland and Nelson 2003). This selectivity could alter not only species diversity and richness, but also

community composition, with less preferred species attaining higher densities in areas accessible to *O. virginianus*.

Although independent effects of *L. maackii* and *O. virginianus* on native plants have been quantified, two studies that measured combined impacts of these species arrived at different conclusions. Gorchov and Trisel (2003) demonstrated that removal of *L. maackii* shoots increased total biomass and stem length of sugar maple (*Acer saccharum*) seedlings that were protected from grazing by *O. virginianus*. However, when unprotected from *O. virginianus*, shoot removal reduced total biomass, root:shoot ratio, and leaf area of seedlings. In addition, biomass, stem length, and leaf area of *Q. rubra* seedlings were greater when roots of *L. maackii* had been severed, but above-ground shoots of *L. maackii* remained. Gorchov and Trisel (2003) attributed these responses to protection of seedlings by *L. maackii* from grazing by *O. virginianus*. These data suggest that management of both *L. maackii* and *O. virginianus* is necessary for successful restoration of *Acer saccharum* and *Quercus rubra*. In contrast, Hartman and McCarthy (2004) found that survival of seedlings of *Q. muehlenbergii* (Chinkapin oak), *Juglans nigra* (black walnut), *Prunus serotina* (black cherry), *Fraxinus pennsylvanica* (green ash), *Cornus floridana* (flowering dogwood), and *Cerci canadensis* (red bud) was greater where *L. maackii* had been removed compared to areas where it was intact. In this case, seedling survival was not enhanced by protection from herbivory by *O. virginianus* (Hartman and McCarthy 2004), suggesting that deer exclusion was not necessary for restoration of invaded communities.

Despite a scarcity of empirical evidence specifically testing whether synergistic interactions between herbivores and invasive plants affect native plant communities, there is reason to predict that these effects would occur. Both light limitation (Boardman 1977) and herbivory (Marquis

1984) reduce growth and survival of herbaceous and woody plants. Negative effects of herbivores thus may be compounded by reduced light availability. For example, Pierson et al. (1990) demonstrated that recovery of invasive *Bromus tectorum* (cheatgrass) that was defoliated decreased when light availability was reduced, and repeated clipping was more likely to result in death of plants grown in shade compared to full sun. Thus, presence of herbivores and reduction in light availability could have negative impacts that are greater than the sum of their independent effects.

Our objectives were to measure the individual and interactive effects of *O. virginianus* and *L. maackii* on species composition, diversity, and richness of forest floor herbs, and to assess their impact on reproduction and abundance of individual herb species and different herb growth forms. We hypothesized that species diversity and richness, as well as growth and abundance of individual herbaceous species would be significantly reduced in the presence of either *L. maackii* or *O. virginianus*. We also hypothesized that species composition would differ among plots with a history of invasion by *L. maackii* and those that had never been invaded, and among plots accessible to *O. virginianus* and those from which *O. virginianus* had been excluded. Lastly, we hypothesized that shade-sensitive species, such as spring perennials, would be more prone to negative impacts of *L. maackii* and *O. virginianus* than other growth forms.

Methods

Study Site

Our study was conducted at the Cincinnati Nature Center, a 405-hectare nature preserve located in Milford, Ohio, approximately 48 km east of Cincinnati. We surveyed forest floor herbs in a second-growth hardwood forest dominated by beech (*Fagus grandifolia*), Chinquapin oak

(*Quercus muehlenbergii*), red oak (*Quercus rubra*), shagbark hickory (*Carya ovata*), bitternut hickory (*Carya cordiformis*), red maple (*Acer rubrum*), and sugar maple (*Acer saccharum*).

Experimental Design

In April 2005, eighteen 10 x 10-m experimental plots were located on a west-facing slope bisected by a gully (1 to 2 m wide, up to 6 m deep) running east to west (Fig. 1). Twelve plots were located north of the gully where *L. maackii* occurred in dense stands. These plots were arranged in 3 north-south rows of 4 plots each, with at least 10 m between each plot and 25 m between each row. In the center of each plot, we established a 5 x 5-m vegetation subplot, to avoid sampling vegetation adjacent to plot edges. Six plots were located south of the gully where there was no history of *L. maackii* invasion; these plots were arranged in 3 north-south rows of 2 plots each with the same interplot spacing as on the north side of the gully. All plots and subplots were demarcated with PVC pipe. Treatments were randomly assigned to plots in a 3 x 2 factorial design representing *L. maackii* present/absent/removed and *O. virginianus* present/absent. There were three replicates per treatment type. We removed *L. maackii* by cutting each shrub off at the base and covering the exposed stump with 2% glyphosate. *Odocoileus virginianus* were excluded with 2.4-m deer fencing (Benner's Gardens, Conshohocken, PA, www.bennersgardens.com) supported by 2.7-m iron L-posts.

Vegetation surveys

We counted and identified all herbaceous plants to species that were located within each 5 x 5-m vegetation subplot in May and August of 2005, 2006, and 2007 (taxonomy follows Gleason and Cronquist 1991). In spring 2007, we measured stem height and leaf number of 36 randomly-selected ramets of false Solomon's seal (*Maianthemum racemosum*) in each plot to determine the effects of *L. maackii* and *O. virginianus* on an individual plant species. We selected *M.*

racemosum because Frankland and Nelson (2003) reported that this species was heavily grazed by deer, and because this species was one of the most abundant herbs at CNC. Further, Rooney (1997) demonstrated that herbivory by *O. virginianus* reduced size of vegetative shoots, frequency of flowering shoots, and shoot densities of a congener, *M. canadense*. We were not able to measure reproduction of *M. racemosa* because only a single individual flowered during spring and summer 2007 when we sampled this species. Although we also measured stems of *M. racemosa* in summer 2007, we were unable to analyze these data because a drought resulted in insufficient numbers of this species for statistical comparisons.

Data Analysis

We calculated Shannon diversity indices for herbs in each plot in 2005, 2006, and 2007 using PC-ORD, version 4 (MjM Software Design, Glenden Beach, OR, 1999). Species diversity and richness were compared between plot types within each season and year with a full-factorial, fixed-factor ANOVA using JMP, version 5.1.2 (SAS Institute Inc., Cary, NC, 2005). Diversity and richness values met assumptions of ANOVA procedures.

Abundance of each species in each treatment type was not normally distributed and contained many zero values. Therefore, comparisons of species composition among treatments was accomplished by using nonmetric multi-dimensional scaling (NMDS), a non-parametric ordination technique, on untransformed counts using PC-ORD. NMDS is widely accepted for ordination of non-normal data, particularly with data sets containing many zeros (Clark 1993, McCune and Grace 2002). NMDS rank orders differences in species compositions between plot types and then calculates an ordination (i.e., graphical) space in which dissimilarity between treatment types most closely matches that in the actual data. Strength of this correlation between

real and ordination data is measured as stress, with high stress representing low correlation. The final ordination of the actual data is the arrangement of plots that produces the least stress.

Count data for each species in each season was analyzed separately for outliers (> 2 STD above mean; PC-ORD outlier analysis). Species identified as outliers or that occurred in fewer than 3 experimental plots were excluded from further analyses. For NMDS tests, we chose the slow and thorough setting on autopilot mode of PC-ORD. This procedure uses 40 runs with real data, 50 runs with randomized data, a maximum of 400 iterations, an instability criterion of 0.00001, and 6 starting axes to select the best dimensionality (McCune and Grace 2002). The ordination was rerun using that dimensionality and the specified starting configuration with no step-down in dimensionality and one run with real data. Sørensen distance measurements were used for all NMDS analyses.

Statistical significance of ordination groupings from NMDS was determined with Multiple Response Permutation Procedures (MRPP), a nonparametric test of equitability between two or more groups. MRPP provides an A statistic of chance-corrected, within-group agreement (McCune and Grace 2002). This procedure allowed us to determine differences in species composition between the 6 treatments. Our data met all assumptions of MRPP such as independence of sample units and use of distance measures that are appropriate to the variable of interest (e.g., Sørensen for community data; McCune and Grace 2002).

We supplemented NMDS and MRPP analyses with Indicator Species Analysis (ISA, in PC-ORD), to identify individual species that were indicative of particular treatments. ISA is robust for data sets that are not normally distributed or contain many tied zeros (Mouillot et al. 2002). ISA assigns Indicator Values (IV) to each species by multiplying relative frequency of each species in each treatment type by the relative abundance of that species. This result is then

multiplied by 100 to generate values ranging from 0 to 100, with 100 indicating that a species is perfectly indicative of a particular treatment. IV's were analyzed for significance using Monte Carlo tests with 1000 randomizations (McCune and Grace 2002).

To determine effects of *L. maackii* or *O. virginianus* on different herb growth forms, we classified each species as one of the following herbaceous growth forms (after Hochstedler et al. 2007): annual, biennial, fern, graminoid, spring perennial, summer perennial, or vine. Because these data could not be normalized, we assigned a rank to each species, with highest abundances assigned to highest ranks. For species with equal abundances, we averaged the consecutive ranks they would have been assigned had they not been equal. We then performed an ANOVA on these average-ranked data. This method approximates a Kruskal –Wallis test, but also provides tests of interactions between factors (Conover and Iman 1981). Similarly, stem height and leaf number of *M. racemosa* could not be normalized, so we performed ANOVA on average-ranked data.

Lastly, to determine effects of *L. maackii* and *O. virginianus* on individual herb species, we focused on rosy sedge (*Carex rosea*), black snakeroot (*Sanicula gregaria*), and common blue violet (*Viola sororia*), the species that were most abundant in each season and year. Because abundance data for these species could not be normalized, we average-ranked counts of each species and performed ANOVA on the average-ranked data. For all multiple comparisons tests, we used Tukey HSD post-hoc tests ($p < 0.05$), which is compatible with ANOVA on average-ranked data (Conover and Iman 1981).

Results

Plant species diversity, richness, and composition

Herb layer species diversity did not differ among treatments in any season of any year (Fig. 2, Appendix 1). There was a significant effect of honeysuckle on species richness in spring ($F_{2,12}$ = 4.00, $p < 0.05$) and summer ($F_{2,12}$ = 4.00, $p < 0.05$) 2007. During these seasons, Tukey HSD revealed that richness was greater in plots where honeysuckle had been removed compared to where it was present, but richness in uninvaded and *L. maackii* present plots was similar in these seasons. In summer 2006, *L. maackii* also was found to be a significant factor ($F_{2,12}$ = 4.21, $p < 0.04$), but richness in this season was lower in plots where honeysuckle was present compared to plots from which it had been removed or had never invaded (Fig. 3). Richness in *L. maackii* removal and uninvaded plots was similar in this season. Presence of *O. virginianus* did not affect species richness, and there was no *O. virginianus* x *L. maackii* interaction (Appendix 2).

Species composition differed among treatments only in Spring 2006 (MRPP, A = 0.0916, $p < 0.04$); plots without a history of invasion by *L. maackii* differed in species composition from all other treatments, but composition was similar in plots from which *O. virginianus* had been excluded and plots where they were present (Fig. 4). ISA revealed that two spring perennials, *Stellaria pubera* (IV = 75, $p < 0.03$) and *Geum canadense* (IV = 55.1, $p < 0.04$), and two annuals, *Galium aparine* (IV = 40.4, $p < 0.03$) and *Chaerophyllum procumbens* (IV = 61.3, $p < 0.03$), were indicative of plots with no history of *L. maackii* invasion with *O. virginianus* excluded. *Eupatorium purpureum* (IV = 91.3, $p < 0.05$), a summer perennial, was indicative of plots with *L. maackii* removed and accessible to *O. virginianus.*

Growth Form Effects

There were no significant interactions between *O. virginianus* and *L. maackii* on abundance of any growth form in any season or year. In spring 2005, abundance of biennials was greater in plots that were accessible to *O. virginianus* ($F_{1,12} = 4.85$, $p = 0.048$; Appendix 3), but abundance of vines was greater in plots from which deer were excluded ($F_{1,12} = 4.85$, $p = 0.048$). In summer 2005, vines were more abundant in plots from which *O. virginianus* had been excluded ($F_{1,12} = 6.60$, $p = 0.02$). Similarly, in spring 2007, abundance of annuals was lower in plots accessible to *O. virginianus* ($F_{1,12} = 9.73$, $p = 0.01$). Presence of *O. virginianus* had no effect on abundances of any other herb growth form.

Honeysuckle treatment significantly did not equally affect the abundance of different herb growth forms (Table 1, Appendix 3). Abundance was greater for both annuals and spring perennials in plots with no history of *L. maackii* invasion compared to plots where it was present for all sample periods except spring and summer 2005 for annuals and summer 2005 and 2007 for spring perennials (Table 1; Appendix 3). In spring of each year, abundance of spring perennials in uninvaded plots tended to be higher than in plots where *L. maackii* was present or had been removed (Fig. 5), although these effects were not always significant (Table 1; Appendix 3).

Presence of *L. maackii* had isolated effects on abundance of ferns, summer perennials, and graminoids. In spring 2006, abundance of summer perennials also was affected by honeysuckle treatment ($F_{2,12} = 3.91$, $p = 0.049$), with greater abundance in plots from which *L. maackii* had been excluded compared to plots where it was present. In spring 2007, abundance of ferns was significantly affected by honeysuckle treatment ($F_{2,12} = 4.17$, $p = 0.04$), with more ferns in uninvaded plots compared to plots where *L. maackii* was present. Honeysuckle treatment

also significantly affected the abundance of graminoids in summer 2007 ($F_{2,12} = 3.92$, $p = 0.049$); graminoid abundance was similar in plots from which *L. maackii* had been removed or had never invaded, but abundances in these plot types was greater than in plots where *L. maackii* was present.

Three Most Abundant Herb Species

Interactions between *L. maackii* and *O. virginianus* did not have a significant effect on abundance of these herb species during any season of any year. Abundance of *V. sororia* in spring 2005 was significantly lower in plots accessible to deer ($F_{1,12} = 5.51$, $p = 0.04$), but *L. maackii* did not affect abundance of *V. sororia* (Appendix 4). Abundance of *S. gregaria* was not affected by *O. virginianus*, but was significantly affected by *L. maackii* treatment; abundance was higher in plots where *L. maackii* had been removed compared to plots where it was present, except in summer 2005 and 2006 (Table 2). There were no effects of *L. maackii* or *O. virginianus* on *C. rosea.*

Effects on *Maianthemum racemosa*

The number of *M. racemosa* leaves was reduced in plots accessible to *O. virginianus* ($F_{1,614} = 338.72$, $p < 0.001$). *Lonicera maackii* treatment also significantly affected leaf number ($F_{2,614} = 12.31$, $p < 0.001$); there were more leaves per ramet in *L. maackii* uninvaded plots than in plots in which *L. maackii* was present. This effect, however, was only evident when *O. virginianus* was excluded. When *O. virginianus* was present, *L. maackii* had no effect on leaf number, as reflected by the significant *L. maackii* x *O. virginianus* interaction ($F_{2,614} = 20.37$, $p < 0.001$; Fig. 6).

Similarly, stem height of *M. racemosa* was greater in plots protected from *O. virginianus* ($F_{1,614} = 434.84$, $p < 0.001$). Effects of *L. maackii* on stem height were dependent upon whether

O. virginianus was present or excluded, as suggested by the significant *L. maackii* x *O. virginianus* interaction ($F_{2,614} = 18.94$, $p < 0.001$). When *O. virginianus* was excluded, stem height was greater in *L. maackii* uninvaded plots than in plots where it had been removed or was present, and greater in plots from which *L. maackii* had been removed than in plots where it was present. However, when *O. virginianus* was present, stem height was greater in plots containing *L. maackii* than in plots from which it had been removed ($F_{2,614} = 10.16$, $p < 0.001$), but there was no difference in stem height in *L. maackii* uninvaded and removed plots (Fig.6).

Discussion

Our study demonstrated that species composition, richness, abundance, and growth of forest floor herbs were altered by *L. maackii* and *O. virginianus*. We also found that these effects were variable across seasons, years, and different levels of organization.

During spring 2006, differences in species composition of herbs were driven by history of invasion by *L. maackii*. Because we removed *L. maackii* in April 2005, the herb community in *L. maackii* removal plots did not have sufficient time to re-establish by spring 2006. Spring perennial herbs typically are adapted to high-light conditions and are less shade tolerant than summer perennials because they grow before canopy trees have leafed out (Sparling 1967, Hughs 1992). Thus, given the early leaf phenology of *L. maackii* (Trisel 1997), spring perennials were more sensitive to shading by *L. maackii* than summer perennials, contributing to altered species composition in spring, but not summer. This is further supported by the negative impact of *L. maackii* on abundance of spring perennials. During spring 2006, the spring perennials *Geum canadense* and *Stellaria pubera* were indicative of plots with no *L. maackii* and no *O. virginianus*; both these species are sensitive to reduced light availability (Hughs 1992).

The annuals *Galium aparine and Chaerophyllum procumbens* also were indicative of plots with no *L. maackii* and no *O. virginianus*, and also have lower reproduction in shaded conditions (Baskin et al. 2004), with survival and fecundity of *G. aparine* specifically reduced under stands of *L. maackii* (Gould and Gorchov 2000). *Stellaria pubera*, an exotic invasive herb, was indicative of plots with no *O. virginianus*, suggesting that biological resistance (i.e., deer herbivory) may reduce its success. These data suggest that native shade-intolerant species, particularly spring perennials and shade-intolerant annuals, are particularly impacted negatively by invasion of *L. maackii*, and conservation of shade-intolerant endangered or threatened species should be of high priority.

In spring 2006, *Eupatorium purpureum* was indicative of plots where *L. maackii* had been removed and were accessible to *O. virginianus*. When eaten, *Eupatorium* species cause an array of potentially fatal conditions in ungulates, livestock, and small mammals (Sharma et al. 1998), linked in part to toxic levels of nitrates (Sund et al. 1957). *Eupatorium purpureum* is a summer perennial, a growth form that may be less sensitive to *L. maackii* (Miller and Gorchov 2004) because it grows after the native canopy has leafed out, and is thus adapted to low-light conditions. Luken et al. (1997) removed *L. maackii* from the understory of a deciduous forest and reported that *E. rugosum* grew only in gaps created from removal of *L. maackii*. Our results suggest that *E. purpureum* also responded to increased light in small gaps in *L. maackii* thickets. Because of the toxic effects of *E. purpureum* on herbivores, it could be considered as a candidate species for re-introduction into areas after removal of *L. maackii* where browsing by *O. virginianus* is a concern.

In summer 2006, richness was reduced in plots from which *L. maackii* was removed. This result could be due to changes in microclimate following shrub removal, and variable rainfall

between years. However, increased species richness in *L. maackii* removal plots in spring and summer 2007 could have been due to increased number of shade-sensitive species. For example, *Polymnia canadensis* and *Sonchus arvensis* were both found exclusively in plots where *L. maackii* had been removed, and growth of both species is known to increase with greater light intensity (Bender et al. 2000, Zollinger and Kells 1991).

In all seasons except summer 2005 and summer 2006, abundance of one of the three most abundant herbs, *S. gregaria*, increased after removal of *L. maackii*, but its abundance did not differ between sites where *L. maackii* was present and sites where it was absent. This finding suggests that *S. gregaria* temporarily responded to an increase in light availability caused by removal of crowns of *L. maackii*, and was not necessarily affected by presence of *L. maackii* shade, per se. Thompson (1980) found that *S. gregaria* quickly dispersed into light gaps from distances > 1 m, because the hooked seedpods of this species facilitates dispersal on animal fur. For studies of invasive plants, these results underline the importance of having un-invaded control sites for comparisons with removal treatments to differentiate between effects due to increased light from the removal of the invasive species, and other direct effects attributable to the invasive species (Gould and Gorchov 2000).

None of the three most abundant herb species were consistently reduced by *O. virginianus* although *V. sororia* was less abundant in plots accessible to *O. virginianus* in spring 2005. The lack of a difference in species diversity or composition between areas with *L. maackii* present and areas where it had been removed suggested that *L. maackii* inhibited re-growth of native vegetation for at least two years post-removal. Such inhibition also was found in the invasive ice plant (*Mesembryanthemum crystallinum*) that increased soil salinity, making restoration of invaded habitats after removal of *M. crystallinum* futile unless these efforts also

were accompanied by soil remediation (El-Ghareeb 1991). Similarly, allelopathic compounds exuded by leaves and stems of *Lonicera maackii* reduced herb growth and germination, but also may promote growth of *L. maackii* seedlings (Trisel 1997, Dorning and Cipollini 2006). While it is not known how long these allelopathic compounds may remain in the soil, they could affect the re-establishment of native vegetation. Stands of *L. maackii* also may reduce diversity of the seed bank (Collier and Vankat 2002), which could slow revegetation of areas after *L. maackii* is removed until they are recolonized from surrounding areas (Vellend 2003).

Presence of *O. virginianus* reduced both stem height and leaf number of *M. racemosa*. Ruhren and Handel (2003) reported increased survival of *M. racemosa* (reported as *Smilacina racemosa*) when protected from herbivory, but no flowering or fruiting even after exclusion of deer. We recorded only a single plant in flower in a deer-excluded plot in May 2007. Frankland and Nelson (2003) reported similar reductions in height and percent of flowering *M. racemosa* in areas grazed by *O. virginianus*. Thus, reproductive success of this species may be dependent upon control of *O. virginianus*.

Grazing by *O. virginianus* may reduce stem height and leaf number of *M. racemosa* to a threshold below which competition with *L. maackii* poses no additional disadvantage. This finding was supported by a lack of an effect of *L. maackii* in the presence of *O. virginianus*. In fact, when *O. virginianus* was present, stem height of *M. racemosa* was greater in plots containing *L. maackii* than in plots from which the shrub had been removed. Thus, compared to plots in which *L. maackii* was present, *L. maackii* had a positive effect on stem height and leaf number of *M. racemosa* when *O. virginianus* was present, but a negative effect when *O. virginianus* was excluded. Apparently, removal of *L. maackii* made *M. racemosa* more accessible to grazing by *O. virginianus*. There were fewer native shrubs in plots with a history of

L. maackii invasion (both *L. maackii* present and removed) than in plots with no history of invasion (personal observation). Thus, with a reduced shrub layer after *L. maackii* removal, and with fewer native shrubs, individual *M. racemosa* in plots where *L. maackii* was removed could have been more susceptible to grazing by *O. virginianus*.

Although *O. virginianus* is a dietary generalist and our data demonstrated little effect of *O. virginianus* on individual growth forms, *O. virginianus* also is capable of selecting specific herbs, as demonstrated with *M. racemosa*. This is particularly true during spring and summer when food is not limiting (Smith 1991). Such concentrated foraging on a few species is in contrast with the more generalized, competitive impacts of *L. maackii*. For example, changes in community composition appeared to be primarily driven by *L. maackii* invasion history. Changes in community composition or species diversity caused by selective feeding of *O. virginianus*, however, could be masked by the more generalized impacts from *L. maackii*. These results demonstrated that indices used to estimate intensity of browsing by *O. virginianus* based on impact to a single species, such as those proposed for sugar maple (Frelich and Lorimer 1985) or *Trillium grandiflorum* (Anderson 1994), may not be appropriate in areas impacted by both *L. maackii* and *O. virginianus* because the impacts of *O. virginianus* may be hidden by impacts of *L. maackii*.

Community composition in plots with no history of *L. maackii* invasion was similar to that in plots containing *L. maackii* during spring 2005 and 2007, and summer 2005, 2006, and 2007. This result may indicate effects of herbivory by *O. virginianus*, or more specifically effects from the "ghost of herbivory past" (Howe et al. 2002). Before 2005, *O. virginianus* had access to all areas on our study site. Their prolonged over-browsing could have depleted the herb community so that recovery of native vegetation would take longer than the three years of our

study. Continued grazing of *O. virginianus* outside our experimental plots could also contribute to slower recovery by reducing availability of source seeds (Rooney and Waller 2003).

Alternatively, lack of a difference between *O. virginianus* present and excluded plots may indicate a non-linear relationship between browse pressure and plant community structure (Rooney and Waller 2003). In our treatments, *O. virginianus* and *L. maackii* were either present or absent. We cannot, therefore, exclude the possibility that native herb diversity is highest at intermediate levels of browsing by *O. virginianus* or density of *L. maackii.*

Interactions between *L. maackii* and *O. virginianus* only were present at the level of a single species, i.e., leaf number and stem height of *M. racemosa.* In fact, most of the negative impacts were due to presence of *L. maackii*. Except for impacts on *M. racemosa* leaf number and stem height, effects of *O. virginianus* were sporadic (e.g., growth forms) or occurred only in one season (e.g., *V. sororia*). *Lonicera maackii*, on the other hand, had more consistent impacts on annuals and spring and summer perennials, consistently reduced abundance of *S. gregaria*, and reduced leaf number and stem height of *M. racemosa*. Nevertheless, removal of *L. maackii* may encourage browsing by *O. virginianus* on individual plant species, as demonstrated with *M. racemosa.*

Our study also demonstrated the importance of measuring the impacts of both invasive species and overabundant herbivores on native plant species at multiple levels. Effects apparent at the level of a single species, such as those of *M. racemosa* or *S. gregaria*, do not necessarily translate into effects at higher levels, such as community composition or plant diversity. Thus, effects seen at the species level (e.g., reduced flowering of individuals) should not be used as indicators for community-wide conservation efforts. Alternatively, absence of effects on individual species does not indicate that management is unnecessary. Categorizing plant species

into ecologically-based groupings, such as growth forms, however, can reveal effects hidden at the species level. However, if there are threatened, endangered, or other species of special interest, post-removal surveys to assess damage to these species caused by *O. virginianus* after removal of *L. maackii* could avoid unforeseen negative effects. These surveys will not only benefit browse-sensitive species, but also will alert managers when control of *O. virginianus* is necessary. Hence, long-term monitoring after removal of an invasive species is important for land managers to ensure that conservation efforts remain successful (Blossey 1999, Simberloff 2003).

Acknowledgments

We thank T. Culley, E. Maurer, D. Gorchov, and G. Uetz for providing invaluable assistance in our experimental design, implementation, and interpretation of our study. We also thank S. Matter for offering helpful advice on statistical analysis and data interpretation, and M. Becus and D. Conover for assisting with identification of plant species. Lastly, we thank J. Brzyski, E. Dame, G. Klein, and S. Puthoff for being willing and dependable field assistants.

Literature Cited

Anderson, R.C. 1994. Height of white-flowered trillium (*Trillium grandiflorum*) as an index of deer browsing intensity. Ecological Applications 4:104-109.

Baskin, C.C., T.S. Hawkins, and J.M. Baskin. 2004. Ecological life cycle of *Chaerophyllum procumbens* variety *shortii* (Apiaceae), a winter annual of the North American eastern deciduous forest. Journal of the Torrey Botanical Society 131:126-139.

Blossey, B. 1999. Before, during and after: the need for long-term monitoring in invasive plant species management. Biological Invasions 1:301-311.

Cabin, R.J., S.G. Weller, D.H. Lawrence, T.W. Flynn, A.K. Sakai, D. Sandquist, and L.J. Hadway. 2000. Effects of long-term ungulate exclusion and recent alien species control on the preservation and restoration of a Hawaiian tropical dry forest. Conservation Biology 14:439-453.

Case, C.M. and M.J. Crawley. 2000. Effect of interspecific competition and herbivory on the recruitment of an invasive alien plant: *Conyza sumatrensis*. Biological Invasions 2:103-110.

Clarke, K.R. 1993. Non-parametric multivariate analyses of changes in community structure. Australian Journal of Ecology 18:117-143.

Collier, M.H. and J.L. Vankat. 2002. Diminished plant richness and abundance below *Lonicera maackii*, and invasive shrub. The American Midland Naturalist 147:60-71.

Conover, D.G. 2007. Impact of deer on vegetation in Miami Whitewater Forest, Mitchell Memorial Forest and Shawnee Lookout Park. Contractual Report, Hamilton Country Parks, Ohio.

Conover, D.G. and D. R. Geiger. 1993. Glycophosphate controls *L. maackii* in native woodland restoration (Ohio). Restoration and Management Notes 11:2 Winter.

Conover, W.J. and R. L. Iman. 1981. Rank transformations as a bridge between parametric and nonparametric statistics. The American Statistician 35:124-129.

Creed, R.P., and S.P. Sheldon. 1995. Weevils and watermilfoil: did a North American herbivore cause the decline of an exotic plant? Ecological Applications 5:1113-1121.

D'Antonio, C. and L. A. Meyerson. 2002. Exotic plant species as problems and solutions in ecological restoration: a synthesis. Restoration Ecology 10:703-713.

Dorning, M. and D. Cipollini. 2006. Leaf and root extracts of the invasive shrub, *Lonicera maackii*, inhibit seed germination of three herbs with no autotoxic effects. Plant Ecology 184:287-296.

Edwards, G.R., G.W. Bourdot, and M.J. Crawley. 2000. Influence of herbivory, competition and soil fertility on the abundance of *Cirsium arvense* in acid grassland. Journal of Applied Ecology 37:321-334.

El-Ghareeb, R. 1991. Vegetation and soil changes induced by *Mesembryanthemum crystallinum* L. in a Mediterranean desert ecosystem. Journal of Arid Environments 20: 321-330.

Frankland, F. and T. Nelson. 2003. Impacts of white-tailed deer on spring wildflowers in Illinois, USA. Natural Areas Journal 23: 341-348.

Frelich, L.E. and C.G. Lorimer. 1991. Natural disturbance regimes in hemlock-hardwood forests of the upper Great Lakes region. Ecological Monographs 61:145-164.

Gleason, H.A and A. Cronquist. 1991. Manual of the Vascular Plants of Northeastern United States and Adjacent Canada, 2nd edition. New York Botanical Garden. Bronx, New York.

Gorchov, D.L. and D.E. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology 166: 13-24.

Gould, A.M.A. and D.L. Gorchov. 2000. Effects of the exotic invasive shrub *Lonicera maackii*, *L. maackii*, on survival and fecundity of native forest annual herbs. The American Midland Naturalist 144:36-50.

Hartman, K.M. and B.C. McCarthy. 2004. Restoration of a forest understory after the removal of an invasive shrub, *L. maackii* (*Lonicera maackii*). Restoration Ecology 12:154-165.

Hartman, K.M. and B.C. McCarthy. 2007. A dendro-ecological study of forest overstory productivity following the invasion of the non-indigenous shrub *Lonicera maackii*. Applied Vegetation Science 10:3-14.

Healey, W.M. 1997. Influence of deer on structure and composition of oak forests in central Massachusetts. pp. 249-266 *In* McShea, W.J., H.B. Underwood, and J.H. Rappole (Eds.). The Science of Overabundance: Deer Ecology and Population Management. Smithsonian Institution Press. Washington D.C.

Hobbs, R.J. and S. E. Humphries. 1995. An integrated approach to the ecology and management of plant invasions. Conservation Biology 9:761-770.

Hochstedler, W.W., B.S. Slaughter, D.L. Gorchov, L.P. Saunders, and M.H.H. Stevens. 2007. Forest floor plant community response to experimental control of the invasive biennial, *Alliaria petiolata* (garlic mustard). Journal of the Torrey Botanical Society 134:155-165.

Howe, H.F., J.S. Brown, and B. Zorn-Arnold. 2002. A rodent plague on prairie diversity. Ecology Letters 5:30-36.

Hughs, J.W. 1992. Effect of removal of co-occurring species on distribution and abundance of *Erythronium americanum* (Liliaceae), a spring ephemeral. American Journal of Botany 79:1329-1336.

Hutchinson, T.F. and J.L. Vankat. 1997. Invasibility and effects of *L. maackii* in southwestern Ohio forests. Conservation Biology 11: 1117-1124.

Jenkins, M.A., C.R. Webster, and J.H. Rock. 2007. Effects of chronic herbivory and historic land use on population structure of a forest perennial, *Trillium catesbaei.* Applied Vegetation Science 10:441-450.

Kellogg, C.H. and S.D. Bridgham. 2004. Disturbance, herbivory, and propagule dispersal control dominance of an invasive grass. Biological Invasions 6:319-329.

Luken, J.O., L.M. Kuddes, and T.C. Tholemeier. 1997. Response of understory species to gap formation and soil disturbance in *Lonicera maackii* thickets. Restoration Ecology 5:229-235.

Luken, J.O. and J.W. Thieret. 1995 Amur honeysuckle (*Lonicera maackii*; Caprifoliaceae): its ascent, decline, and fall. SIDA 16:479-503.

Maron, J.L. and M. Vila. 2001. When do herbivores affect plant invasion? Evidence for the natural enemies and biotic resistance hypotheses. Oikos 95: 361-373.

McCune, B. and J.B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design. Gleneden Beach, Oregon.

Miller, K.E. and D.L. Gorchov. 2004. The invasive shrub, *Lonicera maackii*, reduces growth and fecundity of perennial forest herbs. Oecologia 139:359-375.

Mouillot, D., J.M. Culioli, and T.D. Chi. 2002. Indicator species analysis as a test of non-random distribution of species in the context of marine protected areas. Environmental Conservation 29:385-390.

Myers, J.A., M. Vellend, S. Gardescu, and P.L. Marks. 2004. Seed dispersal by white-tailed deer: implications for long-distance dispersal, invasion, and migration of plants in eastern North America. Oecologia 139:35-44.

Nixon, C.M., L.P. Hansen, P.A. Brewer, J.E. Chelsvig, T.L. Esker, D. Etter, J.B. Sullivan, R.G. Koerkenmeier, and P.C. Mankin. 2001. Survival of white-tailed deer in intensively farmed areas of Illinois. Canadian Journal of Zoology 79:581-588.

Peck, L.J. and J.E. Stahl. 1997. Deer management techniques employed by the Columbus and Franklin County Park District, Ohio. Wildlife Society Bulletin 25:440-442.

Pimental, D., R. Zuniga, and D. Morrison. 2005. Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecological Economics 52:273-288.

Piper, J.K. 1989 Light, flowering, and fruiting within patches of *Smilacina racemosa* and *Smilacina stellata* (Liliaceae). Bulletin of the Torrey Botanical Club 116:247-257.

Rooney, T.P. 1997. Escaping herbivory: refuge effects on the morphology and shoot demography of the clonal forest herb *Maianthemum canadense*. Journal of the Torrey Botanical Society 124:280-285.

Rooney, T.P., and D.M. Waller. 2003. Direct and indirect effects of white-tailed deer in forest ecosystems. Forest Ecology and Management 181:165-176.

Ruhren, S. and S.N. Handel. 2003. Herbivory constrains survival, reproduction and mutualisms when restoring nine temperate forest herbs. Journal of the Torrey Botanical Society 130:34-42.

Sharma, O.P., R.K. Dawra, N.P. Kurade, and P.D. Sharma. 1998. A review of the toxicosis and biological properties of the genus *Eupatorium*. Natural Toxins 6:1-14.

Simberloff, D. 2003. How much information on population biology is needed to manage introduced species? *Conservation Biology* 17: 83-92.

Smith, W.P. 1991. *Odocoileus virginianus*. The American Society of Mammalogists, Mammalian Species no.388: 1-13.

Sparling, J.H. 1967. Assimilation rates of some woodland herbs in Ontario. Botanical Gazette 128:160-168.

Strole, T.A. and R.C. Anderson.1992. White-tailed deer browsing: species preferences and implications for central Illinois forests. Natural Areas Journal 12:139-144.

Sund, J.M., M.J. Wright, and J. Simon. 1957. Weeds containing nitrates cause abortion in cattle. Agronomy Journal 49:278-279.

Thompson, J.N. 1980. Treefalls and colonization patterns of temperate forest herbs. American Midland Naturalist 104:176-184.

Torgerson, O. and W.R. Porath. 1984. White-tailed populations and habitats of the Midwest oak/hickory forests. Pp.411-426 i*n*: L.K. Halls (*Ed.*) White-Tailed Deer: Ecology and Management. Stackpole Books. Pennsylvania.

Trisel, D.E. 1997. The invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae): factors contributing to its success and its effect on native species. Ph.D. Dissertation, Miami University, Oxford, Ohio.

Vellend, M. 2003. Habitat loss inhibits recovery of plant diversity as forests regrow. *Ecology* 84:1158-1164.

Vilà, M. and C. D'Antonio. 1998. Hybrid vigor for clonal growth in *Carpobrotus* (Aizoaceae) in coastal California. Ecological Applications 8:1196-1205.

Vitousek, P.M. 1990. Biological Invasions and ecosystem processes: towards an integration of population biology and ecosystem studies. Oikos 57:7-13.

Webster, C.R., M.A. Jenkins, and J.H. Rock. 2005. Long-term response of spring flora to chronic herbivory and deer exclusion in Great Smoky Mountains National Park, USA. Biological Conservation 125:297-307.

White. E.M., J.C. Wilson, and A.R. Clarke. 2006. Biotic indirect effect: a neglected concept in invasion biology. Diversity and Distributions 12:443-445.

Yates, E.D., D.F. Levia Jr., and C.L. Williams. 2004. Recruitment of three non-native invasive plants into a fragmented forest in southern Illinois. Forest Ecology and Management 190:119-130.

Zar, J.H. 1999. Biostatistical Analysis, 4th edition. Prentice Hall. New Jersey.

Zavaleta, E.S., R.J. Hobbs, and H.A. Mooney. 2001. Viewing invasive species removal in a whole-ecosystem context. Trends in Ecology and Evolution 16:454-459.

Figure 1: Arrangement of experimental plots; each square represents one 10 x 10-m plot.

Not drawn to scale.

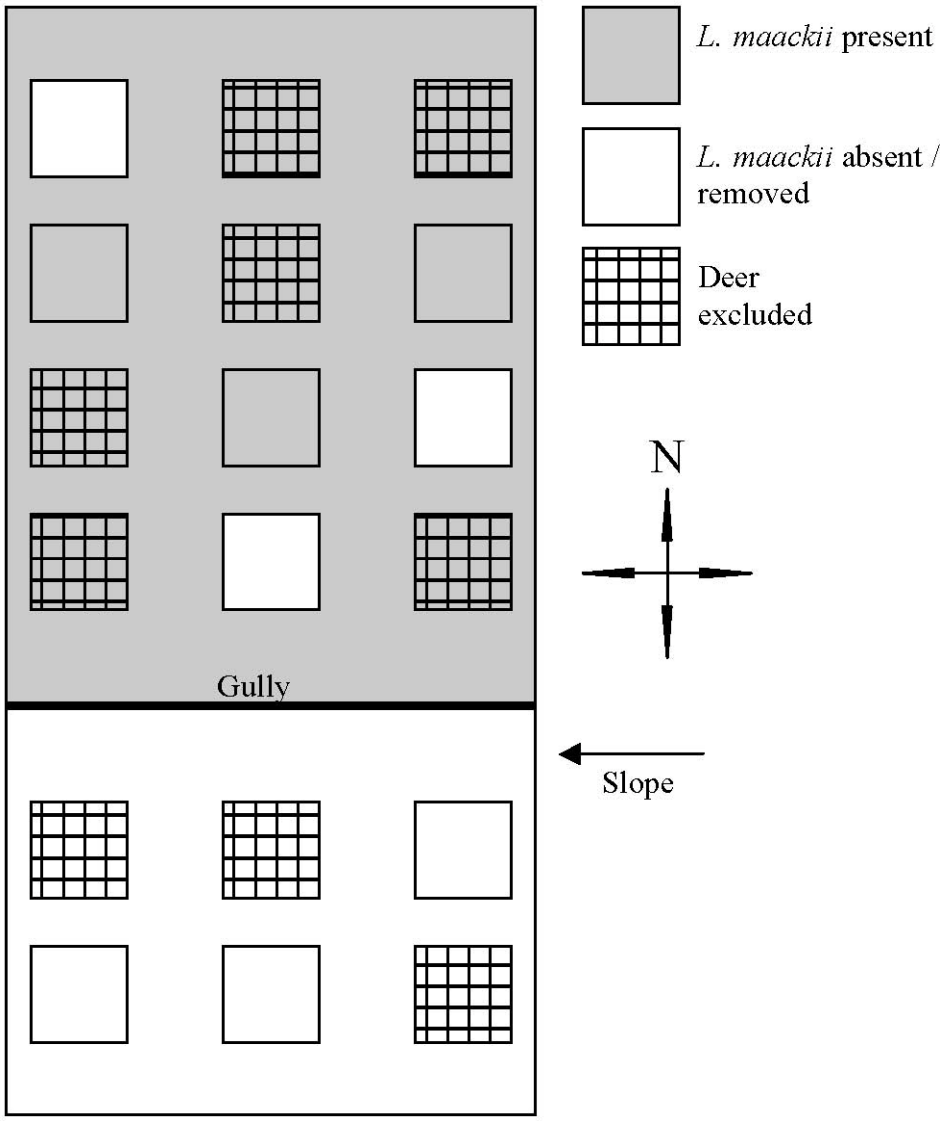

L. maackii present
L. maackii absent / removed
Deer excluded
N
Gully
Slope

Figure 2: Mean diversity (H ± SE) per treatment type for spring and summer 2005, 2006, and 2007

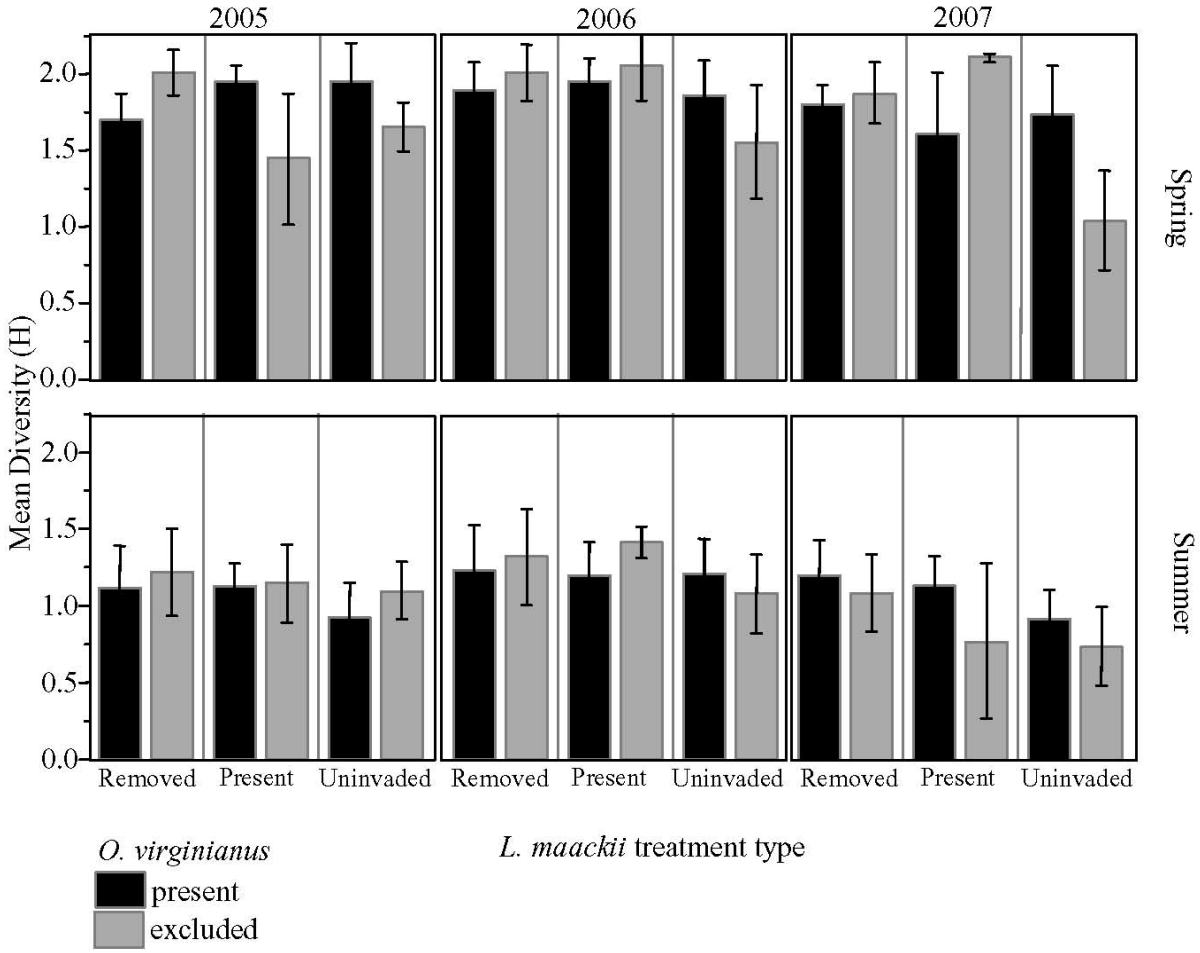

2005
2006
2007
Spring
Summer
Mean Diversity (H)
0.0
0.5
1.0
1.5
2.0
Removed
Present
Uninvaded
L. maackii treatment type
O. virginianus
present
excluded

Figure 3: Mean species richness among *L. maackii* treatment types for spring and summer 2005, 2006, and 2007. Standard error bars are shown. Bars not sharing letters are significantly different at $p < 0.05$. Letters are only shown for comparisons in which a significant difference was detected.

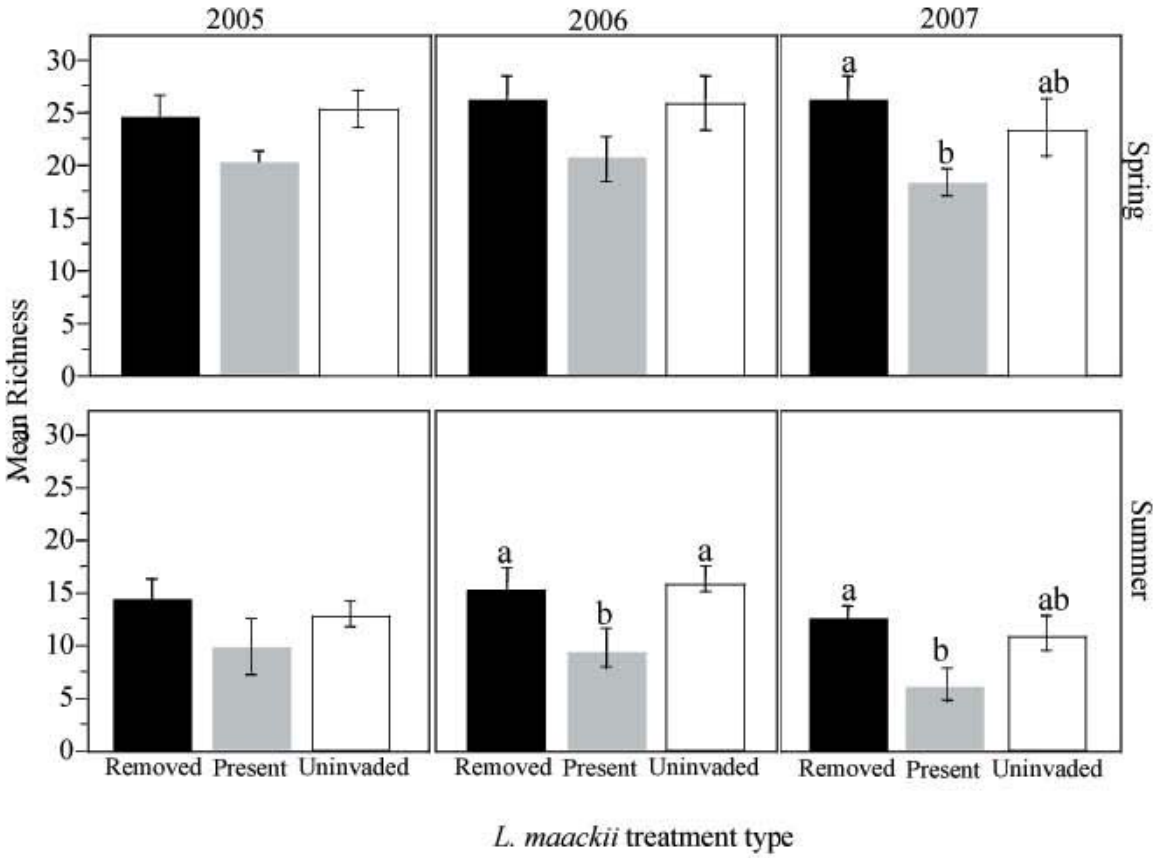

2005
2006
2007
Spring
Summer
Mean Richness
0
5
10
15
20
25
30
a
b
ab
Removed
Present
Uninvaded
L. maackii treatment type

Figure 4: Three-dimensional NMDS ordination of each treatment type in species space for spring 2006, the only season in which MRPP revealed a significant NMDS ordination. Honeysuckle present plots are represented by an inverted triangle, honeysuckle removal plots are represented by an upright triangle, and honeysuckle uninvaded plots are represented by diamonds. Open symbols indicate that deer were excluded, and closed symbols indicate that plots were accessible to deer.

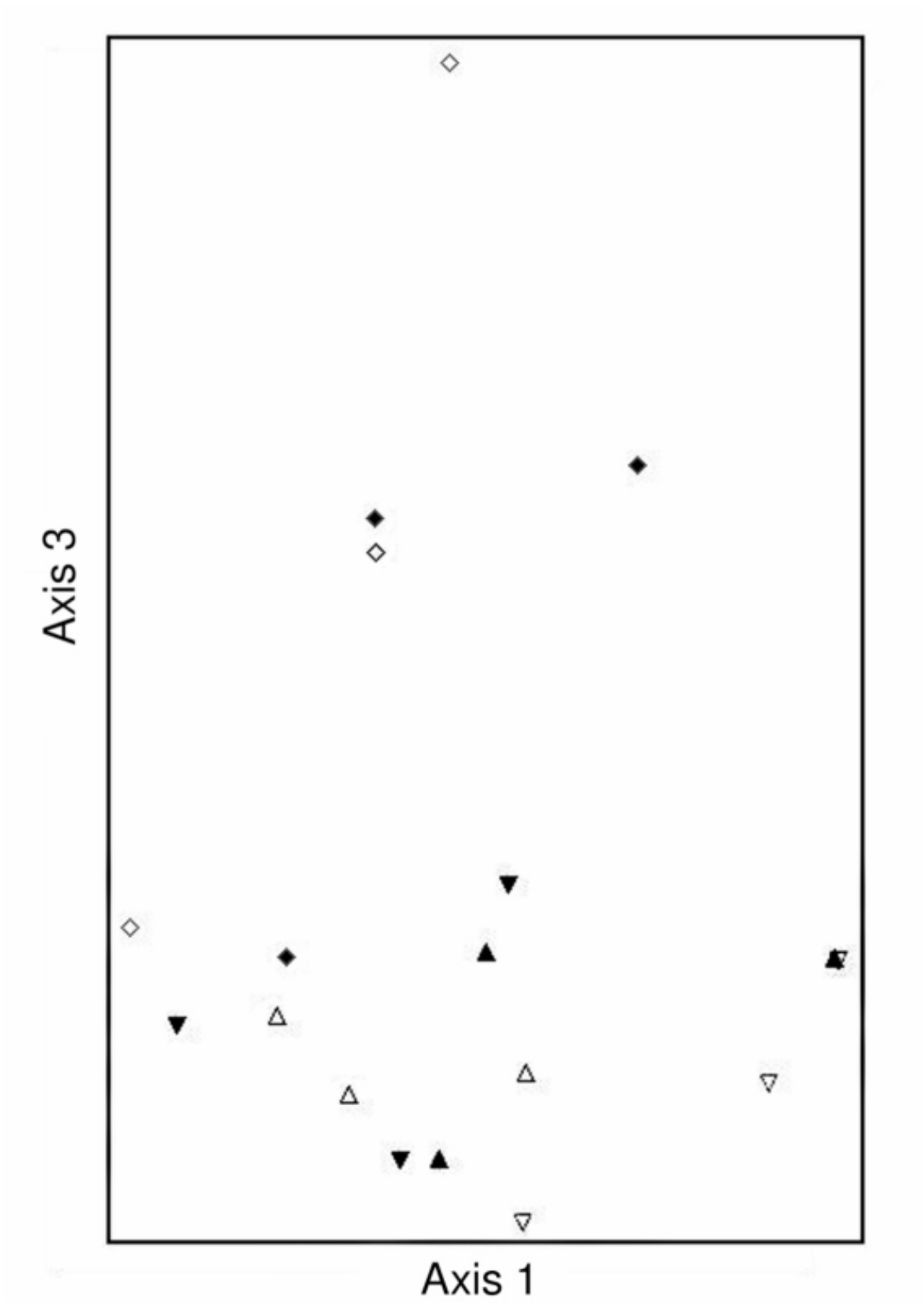

Axis 3
Axis 1

Figure 5: Mean number of spring perennials (± SE) in each *L. maackii* treatment type in spring of 2005, 2006, and 2007.

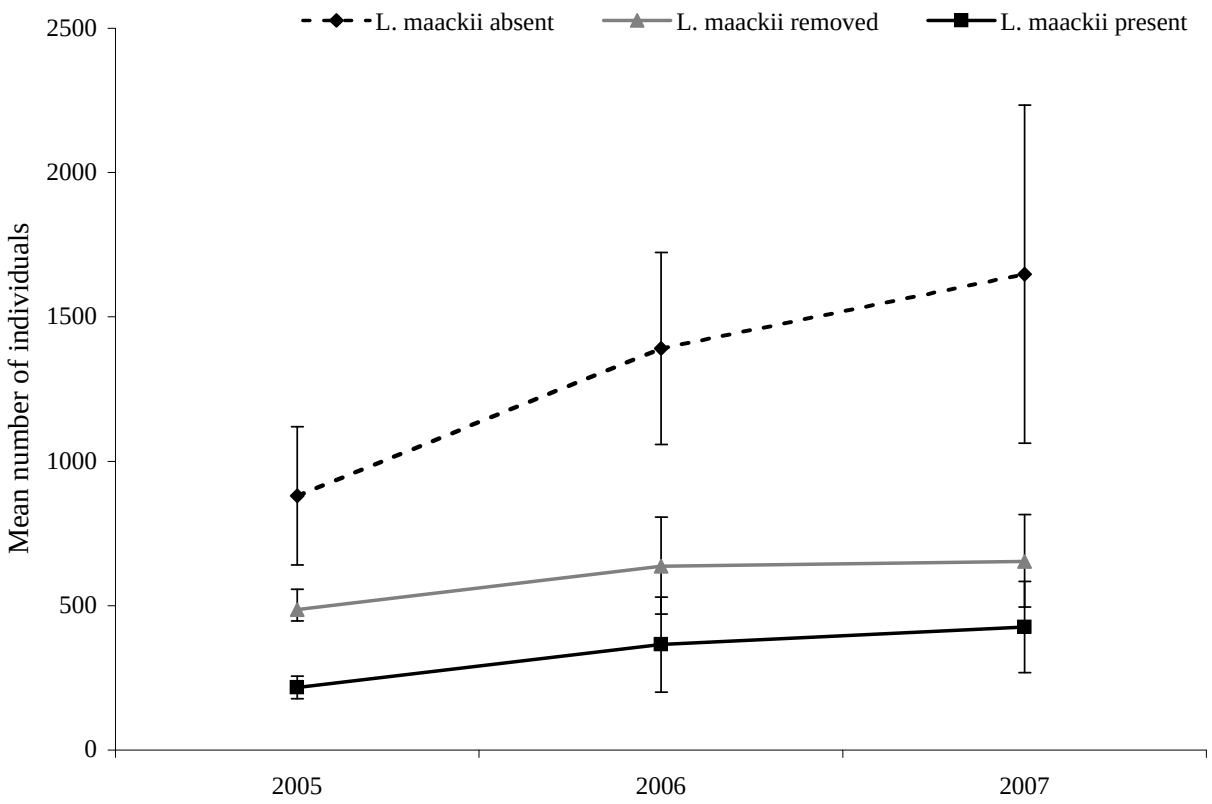

L. maackii absent
L. maackii removed
L. maackii present
Mean number of individuals
2500
2000
1500
1000
500
0
2005
2006
2007

Figure 6: Mean stem height and leaf number (± SE) of *Mainthemum racemosa* in each treatment. Bars sharing letters are not statistically different. Standard error bars are shown.

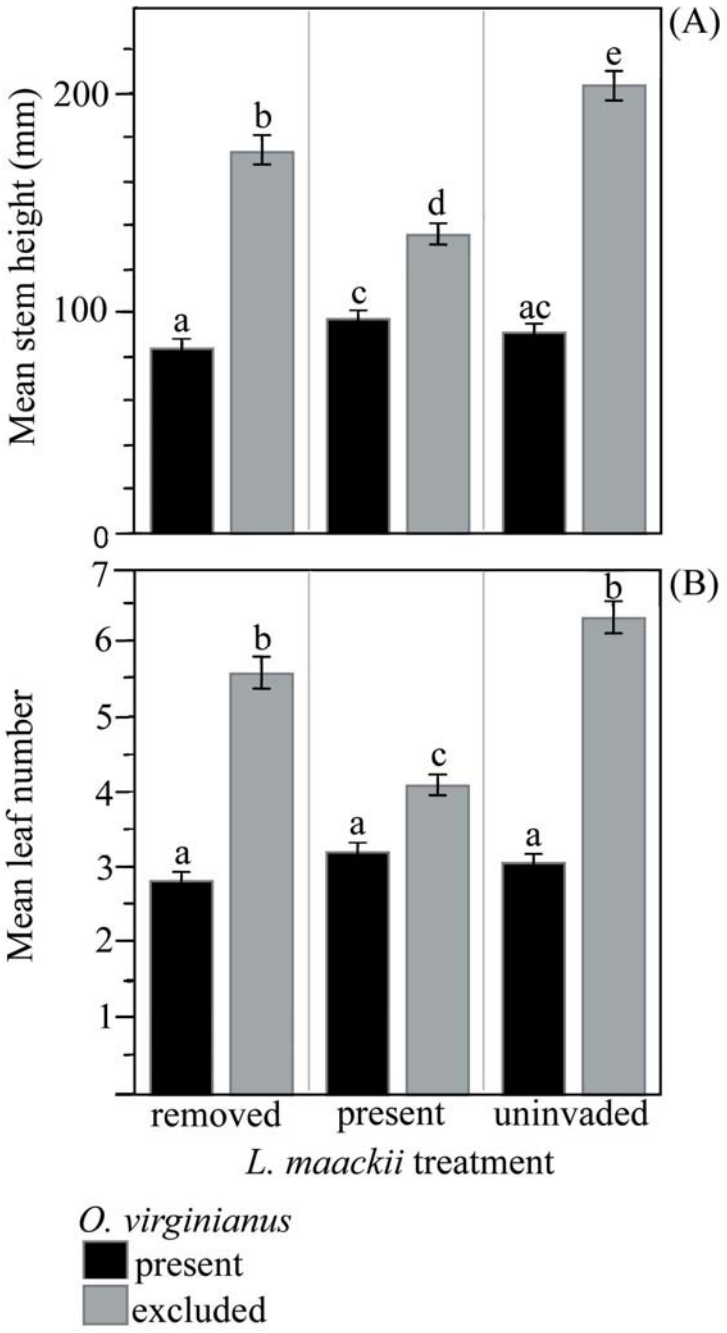

(A)
Mean stem height (mm)
200
100
0
a
b
c
d
ac
e
(B)
Mean leaf number
7
6
5
4
3
2
1
a
b
a
c
a
b
removed
present
uninvaded
L. maackii treatment
O. virginianus
present
excluded

Table 1: Chi square analyses and multiple comparisons results of annual and spring and summer perennial abundance in L. maackii treatments. “NA” indicates that multiple comparisons were not performed because L. maackii treatment types were not significantly different.

		Season	F	df	p	*Lonicera maackii (LM) Multiple* Comparisons
Spring Perennials	**2005**	**Spring**	9.12	2, 12	0.004	*LM present < LM uninvaded* *LM present < LM removed* *LM uninvaded = LM removed*
		Summer	3.36	2, 12	0.069	NA
	2006	**Spring**	12.88	2, 12	0.001	*LM present < LM uninvaded* *LM present < LM removed* *LM uninvaded = LM removed*
		Summer	4.60	2, 12	0.033	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*
	2007	**Spring**	4.40	2, 12	0.037	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*
		Summer	2.67	2, 12	0.110	NA
Annuals	**2005**	**Spring**	0.51	2, 12	0.612	NA
		Summer	1.96	2, 12	0.191	NA
	2006	**Spring**	7.33	2, 12	0.008	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*
		Summer	7.59	2, 12	0.007	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*
	2007	**Spring**	9.12	2, 12	0.004	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*
		Summer	6.91	2, 12	0.010	*LM present < LM uninvaded* *LM present = LM removed* *LM uninvaded = LM removed*

Table 2: Chi square analyses and multiple comparisons results of *Sanicula gregaria* abundance *L. maackii* treatments. "NA" indicates that multiple comparisons were not performed because L. maackii treatment types were not significantly different.

	Season	F	df	p	*L. maackii (LM)* comparisons
2005	**Spring**	4.30	2, 12	0.039	*LM* present = *LM* uninvaded *LM* present < *LM* removed *LM* uninvaded = LM removed
	Summer	0.83	2, 12	0.460	NA
2006	**Spring**	4.91	2, 12	0.028	*LM* present = *LM* uninvaded *LM* present < *LM* removed *LM* uninvaded = LM removed
	Summer	3.14	2, 12	0.080	NA
2007	**Spring**	3.88	2, 12	0.050	*LM* present = *LM* uninvaded *LM* present < *LM* removed *LM* uninvaded = LM removed
	Summer	4.88	2, 12	0.028	*LM* present = *LM* uninvaded *LM* present < *LM* removed *LM* uninvaded = LM removed

Appendix 1: ANOVA tables of effects of white-tailed deer and Amur honeysuckle on diversity of herbaceous vegetation in Spring and Summer 2005, 2006, and 2007.

Spring 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.12709538	0.7579	0.4011
honey	2	0.07678250	0.2289	0.7988
deer*honey	2	0.53366721	1.5912	0.2438
Model	5	0.7375451	0.8796	0.5234
Error	12	2.0123200		
C. Total	17	2.7498650		

Spring 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.00347222	0.0212	0.8867
honey	2	0.29813333	0.9086	0.4291
deer*honey	2	0.16297778	0.4967	0.6205
Model	5	0.4645833	0.5664	0.7245
Error	12	1.9686667		
C. Total	17	2.4332500		

Spring 2007

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.0086366	0.0416	0.8418
honey	2	0.8322290	2.0040	0.1774
deer*honey	2	1.0817092	2.6048	0.1149
Model	5	1.9225749	1.8519	0.1771
Error	12	2.4916494		
C. Total	17	4.4142243		

Summer 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.04073123	0.2464	0.6286
honey	2	0.08248999	0.2495	0.7831
deer*honey	2	0.01850014	0.0560	0.9458
Model	5	0.1417214	0.1715	0.9683
Error	12	1.9834264		
C. Total	17	2.1251477		

Summer 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.01656200	0.0907	0.7684
honey	2	0.08794678	0.2409	0.7896
deer*honey	2	0.08732033	0.2392	0.7909
Model	5	0.1918291	0.2102	0.9517
Error	12	2.1900807		
C. Total	17	2.3819098		

Summer 2007

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.20809313	0.8105	0.3857
honey	2	0.30409900	0.5922	0.5685
deer*honey	2	0.04740218	0.0923	0.9125
Model	5	0.5595943	0.4359	0.8152
Error	12	3.0808083		
C. Total	17	3.6404026		

Appendix 2: ANOVA tables of effects of white-tailed deer and Amur honeysuckle on richness of herbaceous vegetation in Spring and Summer 2005, 2006, and 2007.

Spring 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	14.222222	0.8232	0.3821
honey	2	94.777778	2.7428	0.1045
deer*honey	2	14.777778	0.4277	0.6616
Model	5	123.77778	1.4328	0.2816
Error	12	207.33333		
C. Total	17	331.11111		

Spring 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	1.38889	0.0350	0.8547
honey	2	121.33333	1.5294	0.2561
deer*honey	2	5.77778	0.0728	0.9302
Model	5	128.50000	0.6479	0.6686
Error	12	476.00000		
C. Total	17	604.50000		

Spring 2007

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	56.88889	2.3063	0.1547
honey	2	197.44444	4.0023	0.0466
deer*honey	2	44.11111	0.8941	0.4345
Model	5	298.44444	2.4198	0.0974
Error	12	296.00000		
C. Total	17	594.44444		

Summer 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	37.555556	1.4352	0.2540
honey	2	68.111111	1.3015	0.3079
deer*honey	2	8.777778	0.1677	0.8475
Model	5	114.44444	0.8747	0.5262
Error	12	314.00000		
C. Total	17	428.44444		

Summer 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	10.88889	0.5665	0.4662
honey	2	161.77778	4.2081	0.0412
deer*honey	2	7.11111	0.1850	0.8335
Model	5	179.77778	1.8705	0.1735
Error	12	230.66667		
C. Total	17	410.44444		

Summer 2007

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	3.55556	0.2689	0.6135
honey	2	144.44444	5.4622	0.0206
deer*honey	2	3.11111	0.1176	0.8900
Model	5	151.11111	2.2857	0.1118
Error	12	158.66667		
C. Total	17	309.77778		

Chapter Three

Effects of Invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (Odocoileus *virginianus*) on litter-dwelling invertebrate communities

Abstract: Litter-dwelling invertebrates play crucial roles in litter decomposition and nutrient cycling, and changes in their diversity or abundance can affect these important ecosystem services. Previous studies have outlined the separate effects of invasive plants and overabundant ungulates on species diversity, abundance, and community composition of litter-dwelling invertebrates. However, virtually no studies have examined whether interactions between invasive plants and ungulates affect invertebrate communities. We experimentally examined how invasive Amur honeysuckle (*Lonicera maackii*) and grazing by white-tailed deer (*Odocoileus virginianus*) affected diversity, abundance, and community composition of litter-dwelling invertebrates by excluding deer and removing honeysuckle. We also examined whether deer or honeysuckle affected characteristics of the leaf litter layer such as substrate composition and litter depth. We confirmed that deer reduced total invertebrate abundance, and in particular, abundance of Araneae and Hymenoptera. These effects were most likely related to reduced above-ground biomass of herbaceous vegetation, particularly reduced plant height, in areas accessible to deer. Deer did not affect diversity or composition of litter-dwelling invertebrates. Honeysuckle altered composition of the invertebrate community, but removal of the shrub did not reverse this effect within the two years of our study. Effects of honeysuckle on abundance and diversity were inconsistent and temporally variable. Neither deer nor honeysuckle affected composition of litter substrate, mass of leaf fall, or litter depth, and abundance and diversity of litter invertebrates were not related to litter substrate type. Effects of honeysuckle on

communities of litter invertebrates likely depend on seasonal variation in litter temperature and moisture under the honeysuckle canopy, and to changes in temperature and moisture in canopy gaps created by removal of honeysuckle.

Key Words*: Invasive plant, white-tailed deer, litter, invertebrates,* Lonicera maackii, Odocoileus virginianus*, diversity, abundance, community composition*

Introduction

Litter-dwelling invertebrates are important components of ecosystem processes such as litter decomposition and nutrient cycling (Kremen et al. 1993, Kim 1993). Changes in abundance of invertebrates alter chemistry (Hunter et al. 2003) and mass loss (Bradford et al. 2002) of litter during decomposition, ultimately impacting nutrient cycling (Seastedt and Crossley 1984). Thus, understanding factors that affect invertebrate communities is important for ecosystem conservation and restoration.

Diversity and abundance of litter invertebrates are affected by litter quality and microclimate (Badejo et al. 1998), complexity (Bultman and Uetz 1984), and depth (Uetz 1979, Antvogel and Bonn 2001). In addition to these impacts mediated through the litter layer, abundance and diversity of litter invertebrates are also influenced by plant diversity (Murdoch et al. 1972, Strong et al. 1984), structure (Gibson et al. 1992) and height (Kruess and Tscharntke 2002). Thus, disturbances that alter the plant community or litter layer are likely to be important determinants of abundance and diversity of litter-dwelling invertebrates.

Invasion of native systems by exotic plants affects both plant and litter characteristics (Samways et al. 1996), and also can impact litter invertebrates. For example, invasive *Arundo donax* (giant reed) altered litter moisture, increased proportion of bare ground, and reduced food resources for phytophagous insects along streams in San Francisco (Herrera and Dudley 2003).

Similarly, invasive *Tradescantia fluminensis* (small-leaf spiderwort) altered community composition of litter-dwelling invertebrates in forests of New Zealand by increasing litter moisture and providing greater physical structure than native herbs (Standish 2004). Impacts differ, however, depending on species-specific responses to moisture, temperature, and light penetration under canopies of invasive species (Lindsay and French 2006).

Amur honeysuckle (*Lonicera maackii*; hereafter, honeysuckle) is a shrub introduced to North America from Asia in 1896 for use in horticulture (Luken and Thieret 1995). Since its introduction, honeysuckle has invaded more than 24 states in the eastern United States (Trisel 1997). Once established, it reduces richness and abundance of native herbaceous and woody plants (Collier et al. 2002), growth and seed production of native herbs (Gould and Gorchov 2000, Miller and Gorchov 2004), and survival of native tree seedlings (Gorchov and Trisel 2003, Hartman and McCarthy 2004). Honeysuckle also reduces light penetration to the litter layer (Hutchinson and Vankat 1997), lowers complexity of the litter layer (Buddle et al. 2004), and its shallow, extensive root system may reduce water and nutrient availability to native plants (Hutchinson and Vankat 1997). Consequently, honeysuckle has the potential to affect communities of litter-dwelling invertebrates.

Impacts of honeysuckle on native plant and litter communities may be exacerbated by overabundance of white-tailed deer (*Odocoileus virginianus*; hereafter, deer). There is evidence that deer disperse viable seeds of *L. morrowii* (Vellend 2002), but the extent to which deer disperse seeds of *L. maackii*, or whether leaves of *L. maackii* is a significant food source for deer is not known. Browsing by deer decreases plant growth, reproduction, and density (Rooney 1997), and reduces diversity of native herbaceous vegetation (Rooney and Waller 2003), impacts that can last more than 30 years after deer populations have been reduced (Balgooyen and Waller

1995). Further, these impacts on native plants could subsequently affect diversity, abundance, or community composition of litter-dwelling invertebrates (Murdoch et al. 1972, Strong et al. 1984).

Several studies have examined impacts of other cervid species on ground-dwelling invertebrates. For example, moose (*Alces alces*) reduced cover of bilberry (*Vaccinium myrtillus*), which lowered soil moisture and altered composition of carabid beetle communities (Melis et al. 2007). Reduced abundance of invertebrates also has been attributed to grazing by red deer (*Cervus elaphus*; Baines et al. 1994), Sitka black-tailed deer (*Odocoileus hemionus sitkensis*; Allombert et al. 2005), roe deer (*Capreolus capreolus*) and moose (Suominen et al. 1999a). Positive effects also have been reported for moose (Suominen et al. 1999a and b) and caribou (*Rangifer tarandus*; Suominen et al. 2003). Little is known, however, about the effects of white-tailed deer on invertebrate communities, and no study has examined how the combination of deer and an invasive plant species alter communities of litter invertebrates.

To address this, we used a combination of deer exclusion and honeysuckle removal treatments to determine if these species, independently or together, affected diversity, abundance, and community composition of litter-dwelling invertebrates. Because of the sensitivity of litter-dwelling invertebrates to litter depth and complexity (Bultman and Uetz 1984, Antvogel and Bonn 2001), we also evaluated whether depth of leaf litter and composition of the litter layer were impacted by presence of deer and/or honeysuckle. Lastly, we sought to establish whether such changes in litter depth and substrate composition affected diversity or abundance of litter-dwelling invertebrates.

Methods

Study Site

We conducted our experiment in a second-growth hardwood forest at the Cincinnati Nature Center (CNC), a 405-hectare nature preserve in Milford, Ohio. Eighteen experimental plots were placed on a west-facing slope in a hardwood forest dominated by beech (*Fagus grandifolia*), Chinquapin oak (*Quercus muehlenbergii*), red oak (*Quercus rubra*), shagbark hickory (*Carya ovata*), bitternut hickory (*Carya cordiformis*), red maple (*Acer rubrum*), and sugar maple (*Acer saccharum*). The site was bisected by a gully (app. 2 m wide, 6 m deep) running from east to west. The forest north of the gully was heavily invaded by honeysuckle while the forest to the south had no history of honeysuckle invasion.

Experimental Design

In April 2005, we positioned 12 (10 x 10-m) plots in the area north of the gully invaded by honeysuckle, and six plots in the un-invaded area south of the gully, approximately 75 meters from the invaded plots. Treatments were randomly assigned to plots in a 3 x 2 factorial design representing *L. maackii* present/absent/removed and *O. virginianus* present/absent. Deer were excluded with 2.4-m deer fencing (Benner's Gardens, Conshohocken, PA, www.bennersgardens.com) supported by 2.7-m iron L-type steel posts. Honeysuckle shrubs were removed by cutting at the ground and painting the stumps with 2% glyphosate.

Leaf litter invertebrate sampling

Invertebrates in leaf litter were sampled monthly during months of greatest invertebrate abundance and activity (Bultman and Uetz 1984): June, July, August, and September 2005 and 2006. We sampled litter from five arbitrarily chosen areas within each of our experimental plots using a round 0.1-m^2 quadrat. Each quadrat sampler was made of heavy plastic yard edging

approximately 15 cm tall. At each area, we pushed the bottom edge of the quadrat into the ground to prevent invertebrates from crawling under the edge of the quadrat. All litter from inside the quadrat was quickly removed from the quadrat and placed into a sieve (15-mm mesh) to separate large, intact leaves from smaller debris. The smaller sieved debris was placed into a plastic bag for transport to the laboratory. Any invertebrates remaining on the larger, intact leaves or in the quadrat were collected with an aspirator, and placed into 400-ml Whirl-Pak© bags. Sieved litter was placed into Berlese-Tullgren funnels fitted with 5-mm wire mesh filters positioned above beakers containing 250-ml 70% ethanol for 24 hours or until the litter was thoroughly dried. All invertebrates collected in the Berlese-Tullgren funnels were placed into the corresponding Whirl-Pak© bag from the field, and preserved with 70% ethanol. We identified all non-spider specimens to order, and identified spiders to family.

Substrate and Leaf Litter Measurements

Total abundance of invertebrates was higher in June than in July, August, or September 2005. Consequently, we concluded that sampling leaf litter during June would provide the best assessment of how leaf litter affected invertebrate communities. We measured substrate cover in June 2006 at 9 equally-spaced locations in each plot, systematically arranged in a 3 x 3 grid. To examine differences in substrate type among plots, we constructed a 1 x 1-m sampling grid using heavy string laced through a plastic frame to delineate a grid of one hundred 10 x 10-cm squares. This sample grid was placed on the ground at each sample location and the dominant substrate type in each of the 100 squares was classified as: bare ground, leaves, woody debris, rock, vegetation, or mixed. Mixed substrate was ground sparsely covered with small, scattered stones and decomposing woody debris, but without a dominant substrate. This category allowed us to be more conservative when designating a square as bare ground. After recording percent cover

of each substrate type within each sample grid, we selected four arbitrary squares dominated by leaves and measured to the nearest mm the depth from the ground to the top-most leaf, for a total of 36 measurements in each plot.

Because gradients in leaf fall across experimental plots could have altered substrate composition and depth of leaf litter, we also measured leaf fall in each experimental plot. We randomly placed four (50W x 50L x 20H cm) wire mesh baskets in each experimental plot beginning on 20 September 2005 and 2006. We collected, dried, and weighed all leaves that fell into these baskets bi-weekly until there were no overhead leaves visible, after approximately two months.

Statistical Analyses

We calculated Shannon Diversity Indices for the ordinal data taken in each plot (PC-ORD, version 4; MjM Software Design, Glenden Beach, OR, 1999), but could not transform these indices to attain normality. Therefore, within each month sampled, we assigned a rank to each plot according to its diversity index value, with lowest rank assigned to lowest diversity index and using average ranks in cases of ties. This procedure allowed us to compare diversity indices between honeysuckle and deer treatments within individual months. We performed two-way ANOVA on these average-ranked data, a procedure that approximated non-parametric procedures (i.e., Kruskal-Wallis test) and provided tests of interactions (Conover and Iman, 1981).

Non-metric multidimensional scaling (NMDS) was used on untransformed abundance of arthropod orders to determine differences in invertebrate composition between treatment types. NMDS is a method of ordination recommended for data with non-normal distributions and unequal variances, and is effective with data sets containing many zeros (Clarke 1993, McCune

and Grace 2002). For NMDS tests, we used the autopilot mode of PC-ORD, first running the "slow and thorough" setting, which uses 40 runs with real data, 50 randomized runs, 400 maximum iterations, an instability criterion of 0.00001, and 6 starting axes to select the best dimensionality (McCune and Grace 2002). The ordination was then run using the dimensionality suggested in autopilot and the specified starting configuration with no step-down in dimensionality and one run with real data. Sørensen distance measurements were used for all NMDS analyses. Abundance of each order in each season was analyzed for outliers (> 2 standard deviations above mean; PC-ORD outlier analysis) and if selected as an outlier, the species was excluded from analyses. To determine statistical significance of ordinations produced with NMDS, we used Multiple Response Permutation Procedures (MRPP), a nonparametric test of equitability between groups that provides an "A" value representing chance-corrected, within-group agreement (McCune and Grace 2002).

NMDS and MRPP analyses were further supplemented with Indicator Species Analysis (ISA; PC-ORD), a procedure typically used to identify species that are indicative of specific treatment types. ISA considers relative abundance of each species in each treatment as well as occurrence of that order in each treatment type, allowing for habitat assignment of wide-spread species. ISA is a technique that is robust for non-normal data containing many tied zeros (Mouillot et al. 2002). We applied this method to our ordinal-level data.

We used two-way ANOVA (JMP IN, version 5.1.2; SAS Institute Inc., Cary, NC) to determine differences in substrate composition and depth of leaf litter among treatment types. We calculated proportion of total substrate composed of each substrate type, and transformed these data with an arcsine (square root) transformation (Zar 1999) to meet assumptions of ANOVA. Vegetation and exposed root substrate types were omitted from analyses because of

insufficient occurrences (0.20% and 0.06% mean cover, respectively). Measurements of litter depth met assumptions of ANOVA.

To compare differences in leaf fall among treatments, we calculated mean mass of litter collected from the four litter baskets in each plot for each two-week sampling date (n = 4) each year. This provided us with 3 replicate litter weights per treatment per sampling date. These data were log-transformed for normality and compared among treatments using a repeated measures ANOVA. We also compared total annual leaf fall in each treatment per year. These data could not be transformed to achieve normality, therefore we performed Kruskal-Wallis nonparametric analyses on total dry litter mass.

To determine effects of honeysuckle and deer on abundance of the 5 most abundant orders of litter invertebrates (Acari, Araneae, Collembola, Coleoptera, and Hymentoptera), we used two-way ANOVA with months nested within years to compare abundances among treatment types across months and years. We also examined differences in total abundance of all invertebrates. Since abundances contained many true-zero counts and were Poisson distributed, cube root (Araneae), 4^{th} root (Acari) and log (Collembola, Hymenoptera, and total abundance) transformations were necessary to meet assumptions of ANOVA (Zar 1999). However, we were not able to analyze abundance data of Coleoptera because they were highly skewed in some months, most likely a result of periodic peaks in abundance of Staphylinid beetles. We include abundance of Coleoptera in the figures for comparison with other orders.

We also used ANOVA to examine effects of depth of leaf litter and substrate type on abundance of the 5 most abundant orders, total abundance of invertebrates, and invertebrate diversity. Data for substrate type and invertebrate abundance were transformed as described

above to meet assumptions of ANOVA. Tukey HSD ($p < 0.05$) was used for all ANOVA post-hoc pair-wise comparisons.

Results

Diversity

There were no significant interactions of deer and honeysuckle on diversity of invertebrates in any month or year (Appendix 1). Deer significantly increased invertebrate diversity only in August 2005 ($F_{1,12} = 7.16$, $p = 0.02$; Fig.1) with no effects in other sampling periods (Appendix 1). Effects of honeysuckle on invertebrate diversity were inconsistent and temporally variable (Appendix 1). In August 2005, honeysuckle significantly altered invertebrate diversity ($F_{2,12} = 5.76$, $p = 0.02$; Fig. 1); Tukey HSD revealed that diversity was lower in plots uninvaded by honeysuckle than in plots with honeysuckle invaded or removed. Honeysuckle was also a significant factor in September 2005 ($F_{2,12} = 4.08$, $p = 0.04$; Fig. 1), but diversity in uninvaded plots was higher than in honeysuckle removed plots, and honeysuckle invaded and uninvaded plots did not differ. Alternatively, honeysuckle treatment significantly affected diversity in July 2006 (($F_{2,12} = 3.99$, $p = 0.047$; Fig. 1), but there was no difference in diversity between honeysuckle invaded and uninvaded plots, and diversity in uninvaded plots was lower than in honeysuckle removed plots.

Invertebrate community composition

Composition of invertebrate communities differed significantly between treatments in September 2005 ($A = 0.11$, $p = 0.04$; Fig. 2a), June 2006 ($A = 0.15$, $p = 0.01$; Fig. 2b), and September 2006 ($A = 0.08$, $p = 0.03$; Fig. 2c). In September 2005 and June 2006, two-dimension ordinations separated honeysuckle uninvaded plots from honeysuckle invaded and removed plots along Axis 2 (Fig. 2a,b). In September 2006, uninvaded plots were separated from invaded plots, but only

along a single axis (Fig. 2c). During these months, deer appeared to have no affect on composition of invertebrate communities. Indeed, effects of honeysuckle on composition were apparent in both deer present and excluded plots, indicating there was no interaction of deer and honeysuckle on composition of the invertebrate community. However, although community composition differed among honeysuckle treatments, no invertebrate orders were significantly indicative of a particular treatment type.

Abundance

Total invertebrate abundance was greater in 2006 than in 2005 ($F_{1,96} = 3.92$, $p = 0.05$; Fig. 3). There was a significant effect of month[year] on total invertebrate abundance ($F_{1,96} = 12.15$, $p < 0.0001$); Tukey HSD tests revealed that in 2005, total abundance in June and July were greater than September and in 2006, total abundance in June and July was greater than both August and September (Fig. 3). Despite this temporal variation, presence of deer reduced total invertebrate abundance ($F_{1,96} = 10.44$, $p = 0.002$; Fig. 3) without deer x month[year] or deer x year interactions (Appendix 2).

Honeysuckle was a significant factor in total abundance of invertebrates ($F_{2,96} = 4.35$, $p = 0.02$; Fig. 3); Tukey HSD tests showed that total abundance was significantly higher in honeysuckle-removed plots than in uninvaded plots but there was no difference between invaded and uninvaded plots, nor between honeysuckle removed and invaded plots (Fig. 3). A significant honeysuckle x month[year] ($F_{6,96} = 3.03$, $p = 0.001$) interaction showed that effects of honeysuckle were not equivalent across months or years. Tukey HSD pair-wise comparisons of honeysuckle treatments in each month revealed that total invertebrate abundance differed across honeysuckle treatments only in June 2006. In this month, plots with honeysuckle present had greater abundance of invertebrates than plots uninvaded by honeysuckle but not uninvaded plots

(Fig. 3). There were no interactions of deer and honeysuckle on total invertebrate abundance in any month or year.

Five most abundant orders

Abundance of Acari, Collembola, and Hymenoptera varied by month and abundances of Acari and Araneae varied by year (Table 1). Presence of deer significantly reduced abundance of Araneae and Hymenoptera (Fig. 4, Table 1). Again, although Coleoptera were one of the most abundant orders in our plots, the highly skewed distribution of the abundance data of Coleoptera prevented statistical analyses.

Honeysuckle significantly affected abundance of Acari, Araneae, and Collembola (Table 1). Abundance of Araneae was greater in plots that had not been invaded by honeysuckle than in either honeysuckle invaded or removed plots (Table 1, Fig. 4), but a significant month x honeysuckle [year] interaction revealed this effect was inconsistent across months and years. A significant deer x honeysuckle interaction in abundance of Acari suggested that removal of honeysuckle increased abundance of Acari above that in honeysuckle invaded plots only when deer were excluded (Table 1, Fig. 4). Abundance of Acari also was affected by month x deer [year] and month x honeysuckle [year] interactions (Table 1). Collembola were less abundant in plots uninvaded by honeysuckle compared to invaded and removed plots (Fig. 4, Table 1). There was also a significant deer x honeysuckle x year interaction in Collembola and Hymenoptera abundance (Table 1).

Leaf litter and substrate composition

Neither deer, honeysuckle, nor an interaction of deer and honeysuckle altered leaf litter depth (Table 2) or percent cover (Table 3, Appendix 3) of any substrate type. Leaf fall was greater in plots accessible to deer than in plots from which deer had been excluded in both 2005 ($F_{1,12}$ =

7.65, p = 0.02) and 2006 ($F_{1,12}$ = 7.35, p = 0.02; Table 2). Total annual leaf fall was greater in plots accessible to deer than in plots from which deer had been excluded in 2006 (χ^2 = 5.07, df = 1, p = 0.02), but not in 2005 (χ^2 = 3.27, df = 1, p = 0.07).

Honeysuckle did not affect leaf fall in 2005 ($F_{2,12}$ = 0.78, p = 0.48) or 2006 ($F_{1,12}$ = 1.73, p = 0.22). There were also no interactions of deer and honeysuckle on leaf fall in 2005 ($F_{2,12}$ = 0.18, p = 0.83) or 2006 ($F_{1,12}$ = 0.45, p = 0.64). Peak leaf fall was similar across treatments in 2005 (Table 2). In 2006, peak leaf fall in plots with no history of invasion by honeysuckle did not correspond with peak leaf fall in the other plot types (Table 2). Honeysuckle did not affect total annual leaf fall in either 2005 (χ^2 = 0.33, df = 2, p = 0.85) or 2006 (χ^2 = 3.56, df = 2, p = 0.17). There also were no significant effects of leaf litter depth (Appendix 4) or substrate type (Appendix 5) on total abundance or diversity of invertebrates, or on abundances of the top 5 invertebrate orders.

Discussion

In our study, presence of deer reduced abundance of Araneae and Hymenoptera and lowered total abundance of litter invertebrates. Honeysuckle had significant, but inconsistent, effects on invertebrate diversity and altered composition of invertebrate communities. Abundance and diversity of invertebrates were temporally variable, especially in response to honeysuckle treatment.

Reduced abundance of invertebrates in plots accessible to deer in our study could have been a response to lower plant height or reduced soil penetrability in these plots. Within our experimental plots, height and leaf number of one of the most abundant herbs, *Maianthemum racemosa* (false Solomon's seal), was lower in plots accessible to deer (Christopher and Cameron in preparation). By reducing above ground biomass of herbaceous vegetation, deer

indirectly reduce abundance of litter-dwelling invertebrates (Rooney and Waller 2003). Lower plant height leads to lower abundance and diversity of ground-dwelling arthropods, including Araneae (Rypstra and Carter 1995, Kruess and Tscharntke 2002). Lower abundance of Hymenoptera in deer accessible plots in the present study was most likely due to reduced soil penetration. Abundance and diversity of ants (Formicidae: Hymenoptera) is lower in areas of high soil compaction (Watt et al. 2002). Previously, we found lower penetrability of soil in plots accessible to deer (Christopher and Cameron, in preparation), and ants made up 95.2% of Hymenoptera in our samples.

Honeysuckle would appear to be a minor factor determining abundance or diversity of invertebrates; it contributed to increased abundance only in June 2006 and affected diversity in only three of the eight months of our study. However, in two of the three months in which honeysuckle affected diversity, diversity was higher in plots from which honeysuckle had been removed compared to plots in which it had never invaded. In the remaining month, diversity of invertebrates was higher when honeysuckle was present than in uninvaded plots. In these months, removal of honeysuckle may have created habitat edges which invertebrate species of both understory and open areas can utilize. For example, Shure and Phillips (1991) found that arthropods from surrounding forest readily migrated into small canopy gaps (0.016 hectares); they attributed increased richness in canopy gaps to the differences in habitat characteristics, such as light penetration and litter temperature, between the patches and surrounding forest.

Temporal variation in abundance of the most abundant orders of invertebrates prevented a clear understanding of how honeysuckle impacted abundance of individual invertebrate taxa. For example, honeysuckle treatment significantly affected abundance of Acari during 2006, but this effect was due to removal of honeysuckle in some months and to presence of honeysuckle in

others, and therefore may reflect divergent responses of different acarine taxa. As with total abundance of invertebrates, such temporal variation could be related to seasonal changes in litter moisture, temperature, or stage of decomposition, but these variables were beyond the scope of the present study. Because honeysuckle leaves begin to leaf out earlier than native trees and abscise only after native leaves have fallen, and, forest litter under honeysuckle shrubs remain shaded longer than litter in uninvaded areas. Differences in litter microclimate due to light penetration (e.g., temperature, moisture) between invaded and uninvaded sites should therefore be greatest during late autumn, early winter, and early spring when leaves are present on honeysuckle shrubs, but native canopy trees are still bare. However, abundance of litter invertebrates is typically lower in response to cooler temperatures during these seasons, suggesting impacts of honeysuckle on litter-dwelling invertebrates may be offset by seasonal reductions in temperature.

Deer reduced total abundance of litter invertebrates, but they did not significantly influence diversity or community composition. An exception was in August 2005, when diversity of invertebrates was greater in plots accessible to deer, but this isolated effect is unlikely to be ecologically meaningful. Moderate grazing can indirectly increase diversity of invertebrates by increasing plant diversity (Murdoch et al. 1972, Seymour and Dean 1999), but in a related study in our plots, diversity of herbaceous vegetation was similar in plots from which deer had been excluded compared to deer accessible plots (Christopher and Cameron in preparation). Lack of an effect of deer on diversity of the herb layer also could explain why deer had little effect on diversity or community composition of litter-dwelling invertebrates.

In September 2005, June 2006, and September 2006, plots that had a history of being invaded by honeysuckle (i.e., both honeysuckle present and removed plots) had similar

invertebrate compositions, but differed significantly from plots with no history of honeysuckle invasion. Such alterations in the invertebrate community under honeysuckle could be due to altered microclimate (e.g., litter temperature and moisture) created by a dense shrub canopy. We presumed that removal of the honeysuckle shrub canopy would create an understory more closely resembling that of plots with no history of invasion by honeysuckle, and thus predicted that invertebrate assemblages in honeysuckle removed plots would become more similar to uninvaded plots. However, this was not the case. Plots from which honeysuckle had been removed remained more similar to plots in which the honeysuckle canopy was left intact even two years after removal. This residual effect of honeysuckle could be linked to allelopathic impacts of the shrub (Cipollini et al. 2008). For example, reduction of fungal components of the soil due to allelopathy (Rose et al. 1983) might reduce abundance of fungivorous orders (such as Collembola or Acari) and these effects could cascade to higher trophic levels, including predaceous Coleoptera (e.g., Staphylinidae). Although we did not examine this possibility, increased abundance of fungivorous Collembola and Acari has been associated with increased abundance of predatory Coleoptera and Acari (Chen and Wise 1999). However, despite differences in community composition in three months of our study, we did not identify any invertebrate orders that were indicative of any particular plot type, and therefore cannot determine whether particular invertebrate orders were most associated with uninvaded plots.

There was no difference among treatments in substrate composition or depth of leaf litter. Effects of deer on litter, however, are seasonal, with greatest disturbance to litter in autumn and winter, when availability of plant forage is reduced and deer scratch and disturb the leaf litter layer to forage for tree mast (Smith 1991, Rinkes and McCarthy 2007). Although we did not measure depth of leaf litter in autumn or winter, leaf fall was reduced in plots from which deer

were excluded in 2005 and 2006, and total mass of leaf fall was significantly lower in plots from which deer were excluded in autumn 2006. However, this could reflect our experimental design. Since a 2.4-m deer fence surrounded deer exclusion plots, leaves collected in litter traps would necessarily have come from directly above the plot. In deer accessible plots (without a fence), however, litter traps could have caught leaves falling from directly above as well as those blowing into plots from trees that were not overhead. Considering that leaf fall was similar among honeysuckle treatment types, it was not surprising that composition of litter substrate was unaffected by presence of honeysuckle. We observed leaves from overstory trees entangled in the canopy of honeysuckle shrubs, but our results indicate the canopy was not dense enough to prevent leaves from overhead trees reaching the ground.

Despite the sensitivity of litter-dwelling invertebrates to changes in the litter layer in deciduous forests, neither depth of litter nor composition of the substrate affected total invertebrate abundance, diversity of litter invertebrates, or abundance of the top four invertebrate orders. However, since depth of leaf litter and substrate composition were similar among treatments, the range of leaf depth and substrate composition may not have been sufficient to elicit a response in invertebrate assemblages (Bultman and Uetz 1984).

Although we established our experimental plots to examine individual as well as interactive effects of deer and honeysuckle, only the abundance of Acari was affected by a significant deer x honeysuckle interaction in the present study. This lack of interactions indicates that impacts of deer and honeysuckle on litter-dwelling invertebrates are independent. This may be because effects of deer on litter invertebrates were likely mediated through impacts on plant height or soil compaction, while impacts of honeysuckle were most likely due to alterations in microclimate under honeysuckle canopies. Because we did not specifically test effects of

honeysuckle on microclimate, we suggest that future studies focus on how changes in litter microclimate (e.g., temperature, moisture) that accompany plant invasion affect invertebrates inhabiting the forest floor. It is also important to establish whether such changes affect litter decomposition or nutrient cycling that may ultimately impede restoration of native plant communities.

Acknowledgements

We thank T. Culley, E. Maurer, D. Gorchov, and G. Uetz for providing assistance with our experimental design, implementation, and interpretation of our study. We are also grateful to S. Matter for providing statistical advice and assisting with data interpretation and J. Rutledge and G. Uetz for helping with the identification of difficult specimens. Lastly, we extend our appreciation to D. Capella, E. Dame, G. Klein, J. Kleinhans, J. Lawrence, S. Puthoff, and D. Wright for invaluable field and laboratory assistance.

Literature Cited

Allombert, S., S. Stockton, J.L. Martin. 2005. A natural experiment on the impact of overabundant deer on forest invertebrates. Conservation Biology 19:1917-1929.

Antvogel, H. and A. Bonn. 2001. Environmental parameters an microspatial distribution of insects: a case study of carabids in an alluvial forest. Ecography 24:470-482.

Augustine, D.J. and S.J. McNaughton. 1998. Ungulate effects on the functional species composition of plant communities: herbivore selectivity and plant tolerance. Journal of Wildlife Management 62:1165-1183.

Badejo, M.A., T.I. Nathaniel, and G. Tian. 1998. Abundance of springtails (Collembola) under four agroforestry tree species with contrasting litter quality. Biology and Fertility of Soils 27:15-20.

Baines, D., R.B. Sage, M.M. Baines. 1994. The implications of red deer grazing to ground vegetation and invertebrate communities of Scottish native pinewoods. Journal of Applied Ecology 31:776-783.

Balgooyen, C.P. and D.M. Waller. 1995. The use of *Clintonia borealis* and other indicators to gauge impacts of white-tailed deer on plant communities in northern Wisconsin, USA. Natural Areas Journal. 15:308-318.

Bradford, M.A., G.M. Tordoff, T. Eggers, T. Hefin Jones, and J.E. Newington. 2002. Microbiota, fauna, and mesh size interactions in litter decomposition. Oikos 99:317-323.

Buddle, C.M., S. Higgins, and A.L. Rypstra. 2004. Ground-dwelling spider assemblages inhabiting riparian forests and hedgerows in a agricultural landscape. The American Midland Naturalist 151:15-26.

Bultman, T.L. and G.W. Uetz. 1984. Effect of structure and nutritional quality of litter on abundances of litter-dwelling arthropods. The American Midland Naturalist 111:165-172.

Chen, B. and D.H. Wise. 1999. Bottom-up limitation of predaceous arthropods in a detritus-based terrestrial food web. Ecology 80:761-772.

Cipollini, D., R. Stevenson, S. Enright, A. Eyles, and P. Bonello. 2008. Phenolic metabolites in leaves of the invasive shrub, *Lonicera maackii*, and their potential phytotoxic and anti-herbivory effects. Journal of Chemical Ecology 34:144-152.

Clarke, K.R. 1993. Non-parametric multivariate analyses of changes in community structure. Australian Journal of Ecology 18:117-143.

Collier, M.H., J.L. Vankat, and M.R. Hughs. 2002. Diminished plant richness and abundance below *Lonicera maackii*, and invasive shrub. The American Midland Naturalist 147:60-71.

Conover, W.J. and R. L. Iman. 1981. Rank transformations as a bridge between parametric and nonparametric statistics. The American Statistician 35:124-129.

Gibson, C.W.D., V.K. Brown, L. Losito, and G.C. McGavin. 1992. The response of invertebrate assemblies to grazing. Ecography 15:166-176.

Gist, C.S. and D.A. Crossley, Jr. 1975. The litter arthropod community in a southern Appalachian hardwood forest: numbers, biomass and mineral element content. The American Midland Naturalist 93:107-122.

Gorchov, D.L. and D.E. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology 166: 13-24.

Hartman, K.M. and B.C. McCarthy. 2004. Restoration of a forest understory after the removal of an invasive shrub, *L. maackii* (*Lonicera maackii*). Restoration Ecology 12:154-165.

Herrera, A.M. and T.L. Dudley. 2003. Reduction of riparian arthropod abundance and diversity as a consequence of giant reed (*Arundo donax*) invasion. Biological Invasions 5:167-177.

Hunter, M.D., S. Adl, C.M. Pringle, and D.C. Coleman. 2003. Relative effects of macroinvertebrates and habitat on the chemistry of litter during decomposition. Pedobiologia 47:101-115.

Hutchinson, T.F. and J.L. Vankat. 1997. Invasibility and effects of *L. maackii* in southwestern Ohio forests. Conservation Biology 11: 1117-1124.

Kim, K.C. 1993. Biodiversity, conservation and inventory: why insect matter. Biodiversity and Conservation 2:191-214.

Kremen, C., R.K. Colwell, T.L. Erwin, D.D. Murphy, R.F.Noss, and M.A. Sanjayan. 1993. Terrestrial arthropod assemblages: their use in conservation planning. Conservation Biology 7:796-808.

Kruess, A. and T. Tscharntke. 2002. Contrasting responses of plant and insect diversity to variation in grazing intensity. Biological Conservation 106:293-302.

Levings, S.C. and D. M. Windsor. 1984. Litter moisture content as a determinant of litter arthropod distribution and abundance during the dry season on Barro Colorado Island, Panama. Biotropica 16:125-131.

Lindsay, E.A. and K. French. 2006. The impact of the weed *Chrysanthemoides monilifera* spp. *rotundata* on coastal leaf litter invertebrates. Biological Invasions 8:177-192.

Luken, J.O. and J.W. Thieret. 1995 Amur honeysuckle (*Lonicera maackii*; Caprifoliaceae): its ascent, decline, and fall. SIDA 16:479-503.

McCune, B. and J.B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design. Gleneden Beach, Oregon.

Melis, C., M. Sundby, R. Andersen, A. Moksnes, B. Pedersen, and E. Røskaft. 2007. The role of moose *Alces alces* L. in boreal forest – the effect on ground beetles (Coleoptera, Carabidae) abundance and diversity. Biodiversity and Conservation 16:1321-1335.

Miller, K.E. and D.L. Gorchov. 2004. The invasive shrub, *Lonicera maackii*, reduces growth and fecundity of perennial forest herbs. Oecolgia 139:359-375.

Mouillot, D., J.M. Culioli, and T.D. Chi. 2002. Indicator species analysis as a test of non-random distribution of species in the context of marine protected areas. Environmental Conservation 29:385-390.

Murdoch, W.W., F.C. Evans, C.H. Peterson. 1972. Diversity and pattern in plants and insects. Ecology 53:819-829.

Niemela, J., Y. Haila, and P. Punttila. 1996. The importance of small-scale heterogeneity in boreal forests: variation in diversity in forest-floor invertebrates across succession gradient. Ecography 19:352-368.

Rinkes, Z.L. and B.C. McCarthy. 2007. Ground layer heterogeneity and hardwood regeneration in mixed oak forest. Applied Vegetation Science 10:279-284.

Rooney, T.P. 1997. Escaping herbivory: refuge effects on the morphology and shoot demography of the clonal forest herb *Maianthemum canadense*. Journal of the Torrey Botanical Society 124:280-285.

Rooney, T.P. and D.M. Waller. 2003. Direct and indirect effects of white-tailed deer in forest ecosystems. Forest Ecology and Management 181:165-176.

Rose, S. L. , D. A. Perry, D. Pilz and M. M. Schoeneberger. 1983. Allelopathic effects of litter on the growth and colonization of mycorrhizal fungi. Journal of Chemical Ecology 9: 1153-1162.

Rypstra, A.L. and P.E. Carter. 1995. The web-spider community of soybean agroecosystems in southwestern Ohio. Journal of Arachnology 23:135-144.

Samways, M.J., P.M. Caldwell, and R. Osborn. 1996. Ground-dwelling invertebrate assemblages in native, planted, and invasive vegetation in South Africa. Agriculture, Ecosystems, and Environment 59:19-32.

Seastedt, T.R. and D.A. Crossley, Jr. 1984. The influence of arthropods on ecosystems. BioScience 34:157-161.

Seymour, C.L. and W.R.J. Dean. 1999. Effects of heavy grazing on invertebrate assemblages in the Succulent Karoo, South Africa. Journal of Arid Environments 43:267-286.

Smith, W.P. 1991. *Odocoileus virginianus*. The American Society of Mammalogists, Mammalian Species no.388: 1-13.

Standish, R.J. 2004. Impact of an invasive clonal herb on epigaeic invertebrates in forest remnants in New Zealand. Biological Conservation 116:49-58.

Strong, D.R., J.H. Lawton, and R. Southwood. 1984. Insects on Plants, Community Patterns and Mechanisms. Harvard University Press. Cambridge, Massachusetts.

Suominen, O., K. Danell, and R. Bergström. 1999a. Moose, trees, and ground-dwelling invertebrates: indirect interactions in Swedish pine forests. Oikos 84:215-226.

Suominen, O., K. Danell, and J.P. Bryant. 1999b. Indirect effects of mammalian browsers on vegetation and ground-dwelling insects in an Alaskan floodplain. Écoscience 6:505-510.

Suominen, O. J. Niemelä, P. Martikainen, P. Niemelä, and I. Kojola. 2003. Impact of reindeer grazing on ground-dwelling Carabidae and Curculionidae assemblages in Lapland. Ecography 26:503-513.

Trisel, D.E. 1997. The invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae): factors contributing to its success and its effect on native species. Ph.D. Dissertation, Miami University, Oxford, Ohio.

Vellend, M. 2002. A pest and an invader: white-tailed deer (*Odocoileus virginianus* Zimm.) as a seed dispersal agent for honeysuckle shrubs (*Lonicera*). Natural Areas Journal 22:230-234.

Watt, A.D., N. E. Stork, and B. Bolton. 2002. The diversity and abundance of ants in relation to forest disturbance and plantation establishment in southern Cameroon. Journal of Applied Ecology 39:18-30.

Zar, J.H. 1999. Biostatistical Analysis, 4th edition. Prentice Hall. New Jersey.

Figure 1: Mean (± S.E.) Shannon diversity of orders of litter-dwelling invertebrates in 2005 and 2006. Asterisks indicate significant effects of deer and different letters above bars indicate significant honeysuckle effects.

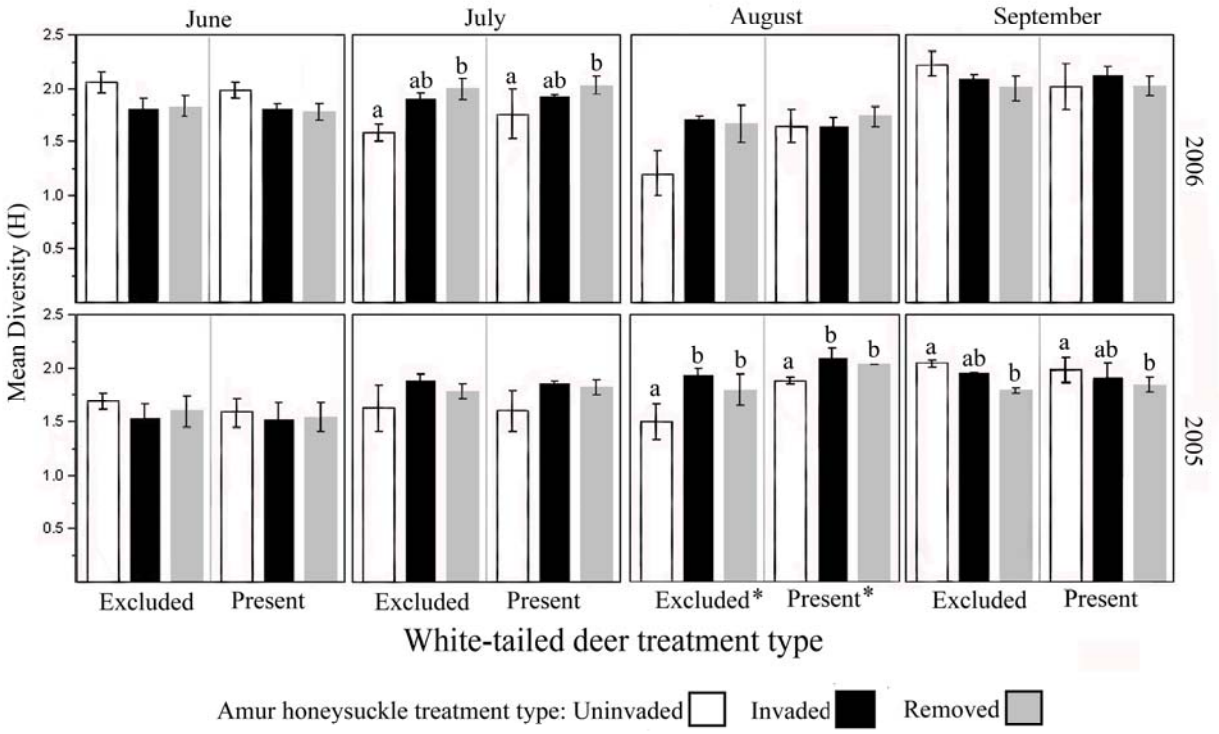
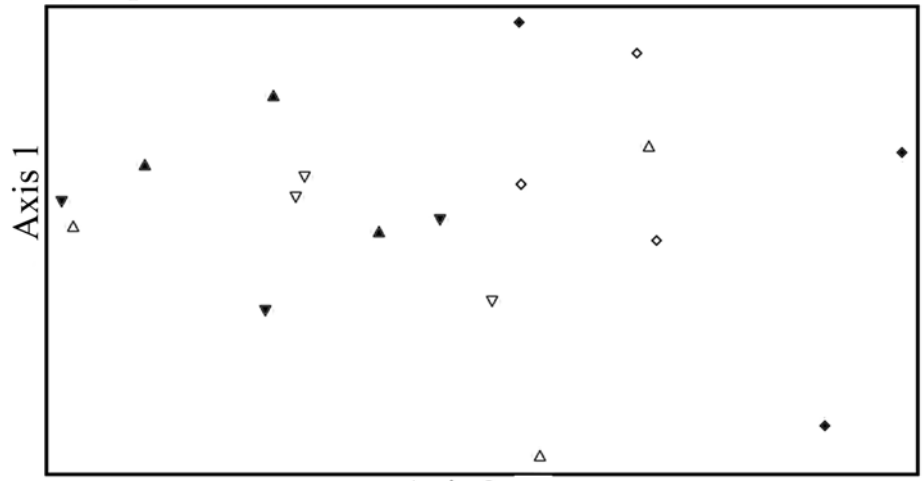
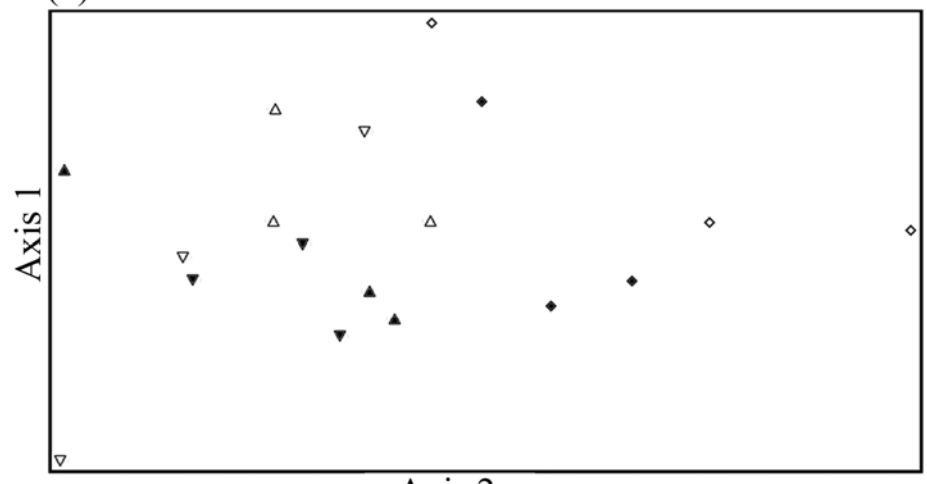

June
July
August
September
2006
2005
Mean Diversity (H)
2.5
2.0
1.5
1.0
0.5
a
ab
b
Excluded
Present
Excluded*
Present*
White-tailed deer treatment type
Amur honeysuckle treatment type: Uninvaded
Invaded
Removed

Figure 2: Non-metric multidimensional scaling ordinations of invertebrate orders. Honeysuckle invaded plots are represented by an inverted triangle, honeysuckle removal plots are represented by an upright triangle, and honeysuckle uninvaded plots are represented by diamonds. Open symbols indicate that deer were excluded, and closed symbols indicate that plots were accessible to deer.

(a) September 2005



(b) June 2006



(C) September 2006

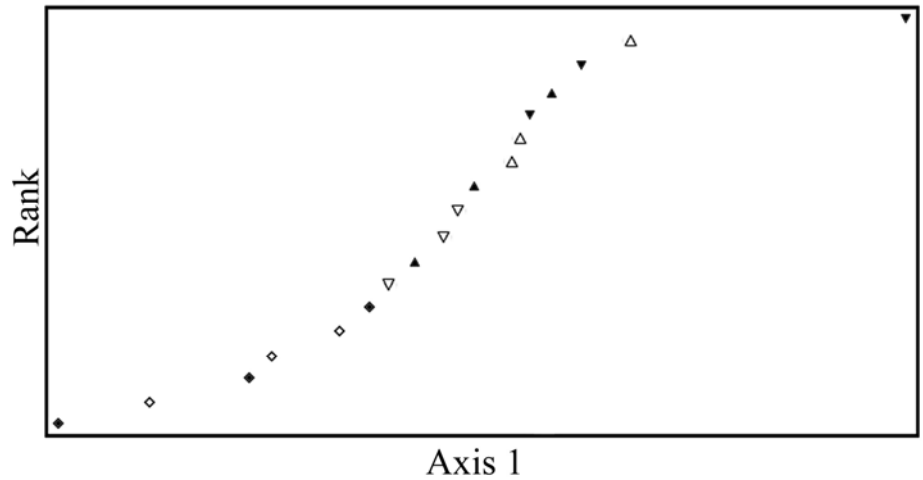


Figure 3: Mean (± S.E.) number of invertebrate individuals / 0.1m^2 quadrat in 2005 and 2006. Different letters above bars indicate significant differences among treatments.

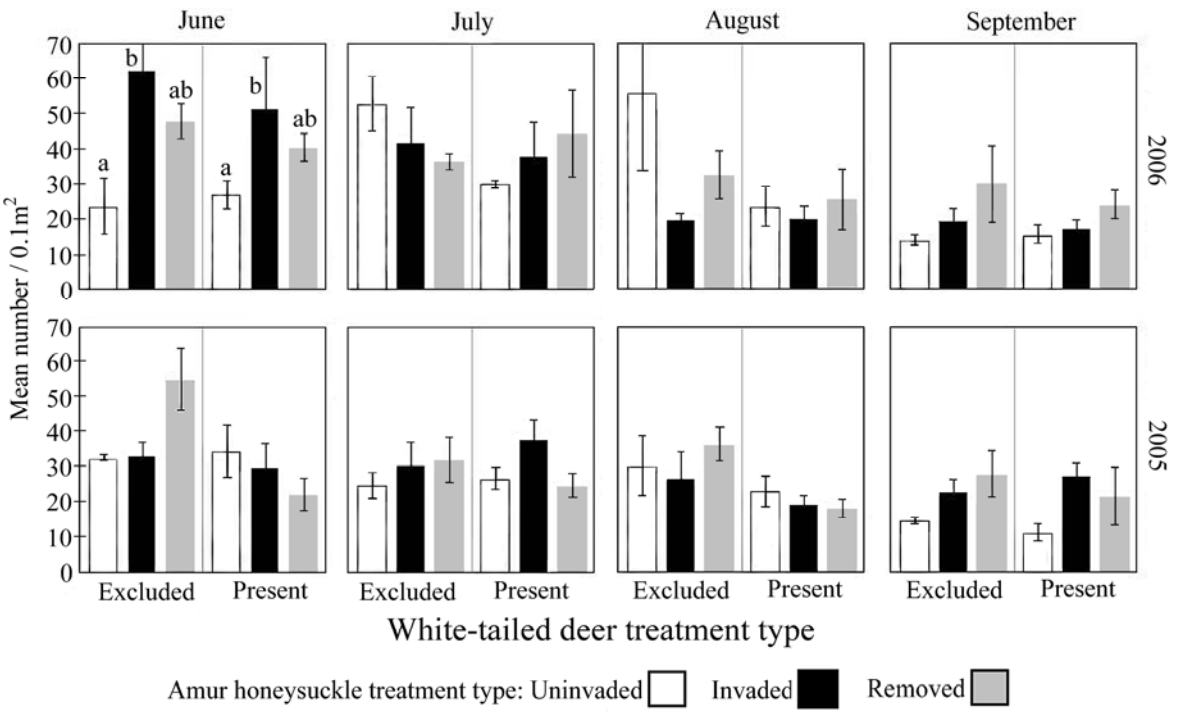

June
July
August
September
2006
2005
Mean number / 0.1m2
a
b
ab
a
b
ab
Excluded
Present
White-tailed deer treatment type
Amur honeysuckle treatment type: Uninvaded
Invaded
Removed

Figure 4: Mean (± S.E.) abundance / 0.1m^2 of the 5 orders of litter-dwelling invertebrates that were numerically most abundant in each treatment type in 2005 and 2006 (pooled).

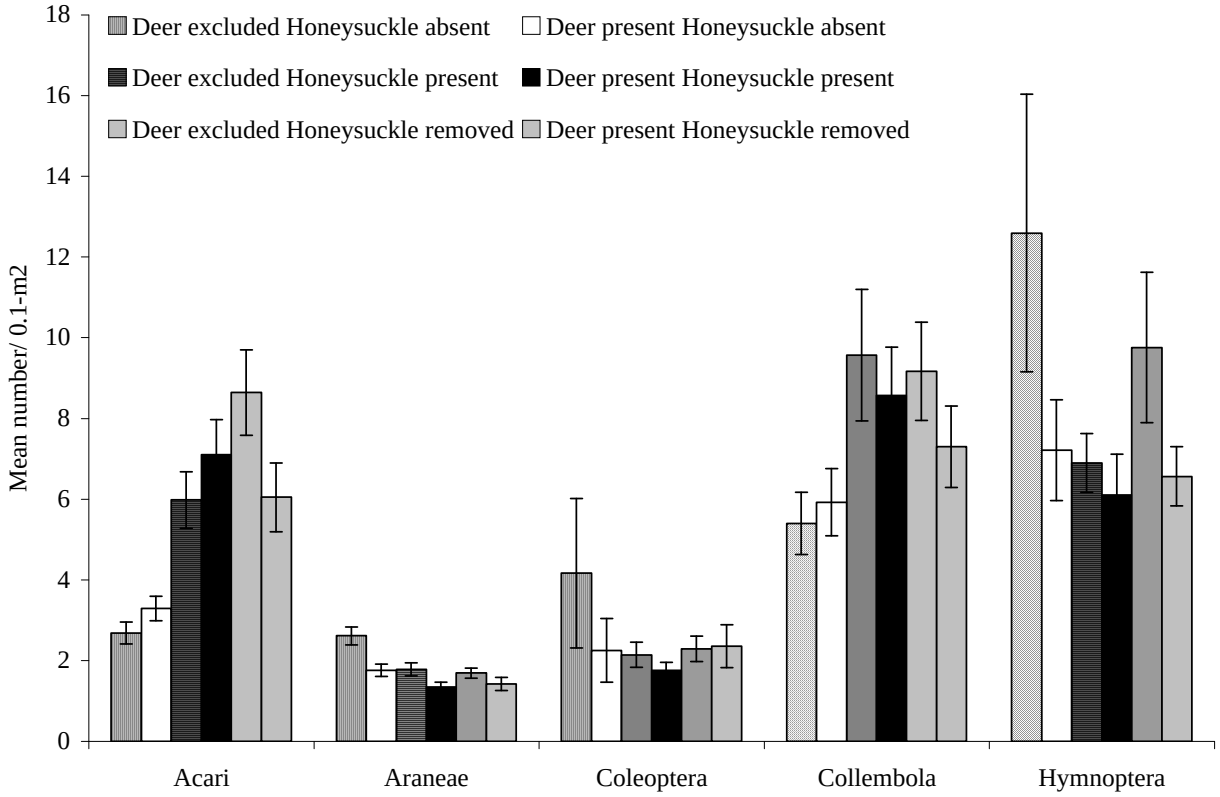

Deer excluded Honeysuckle absent
Deer present Honeysuckle absent
Deer excluded Honeysuckle present
Deer present Honeysuckle present
Deer excluded Honeysuckle removed
Deer present Honeysuckle removed
Mean number/ 0.1-m2
0
2
4
6
8
10
12
14
16
18
Acari
Araneae
Coleoptera
Collembola
Hymnoptera

Table 1: Results of ANOVA of abundances of 5 most abundant invertebrate orders. Bold indicates p-values < 0.05.

	Araneae				Acari			Collembola			Hymenoptera		
Source	DF	SS	F	p	SS	F	p	SS	F	p	SS	F	p
Deer	1, 96	0.50	18.24	**< 0.001**	0.01	0.07	0.795	0.44	1.85	0.178	2.18	4.86	**0.030**
Honeysuckle	2, 96	0.62	11.48	**< 0.001**	2.65	39.10	**< 0.001**	4.35	9.26	**< 0.001**	0.74	0.83	0.441
Deer x Honeysuckle	2, 96	0.06	1.03	0.362	0.41	6.04	**0.003**	0.45	0.96	0.389	0.41	0.45	0.639
Year	1, 96	0.22	7.85	**0.006**	0.38	11.21	**0.001**	0.48	2.05	0.156	0.26	0.57	0.456
Deer x Year	1, 96	0.01	0.21	0.648	0.09	2.40	0.125	0.09	0.36	0.554	0.01	0.01	0.936
Honeysuckle x Year	2, 96	0.09	1.50	0.229	0.33	4.83	**0.010**	0.55	1.17	0.317	0.08	0.08	0.923
Deer x Honeysuckle x Year	2, 96	0.12	2.07	0.132	0.10	1.42	0.247	1.57	3.35	**0.040**	3.11	3.48	**0.035**
Month [Year]	6, 96	0.27	1.63	0.149	1.40	6.87	**< 0.001**	40.91	29.04	**< 0.001**	24.25	9.04	**< 0.001**
Month x Deer [Year]	6, 96	0.19	1.15	0.344	0.46	2.25	**0.046**	0.40	0.28	0.946	2.96	1.11	0.367
Month x Honeysuckle [Year]	12, 96	0.82	2.53	**0.006**	0.77	1.90	**0.044**	5.92	2.10	**0.024**	9.43	1.76	0.067
Month x Deer x Honeysuckle [Year]	12, 96	0.36	1.10	0.369	0.32	0.77	0.680	2.01	0.72	0.737	3.71	0.69	0.758

Table 2: Mean (± S.E.) depth of leaf litter, and mean anual leaf fall in each experimental treatment. Means are based on 3 plots.

Treatments		Leaf Litter	Mean annual leaf fall (dry g / m^2)	
Deer	Honeysuckle	Depth (mm)	2005	2006
Excluded	Uninvaded	31 ± 1.4	355 ± 4.3	376 ± 45.3
Excluded	Invaded	32 ± 1.1	389 ± 13	457 ± 14.5
Excluded	Removed	30 ± 1.6	406 ± 42.3	442 ± 38
Present	Uninvaded	36 ± 1.3	468 ± 32.6	452 ± 9.3
Present	Invaded	28 ± 1.2	421 ± 42	329 ± 45
Present	Removed	30 ± 1.3	462 ± 85	593 ± 58

Table 3: Mean (± S.E.) percent cover of each substrate type per treatment. Means are based on 3 plots.

Treatments		Substrate Categories						
Deer	Honeysuckle	Bare	Rock	Woody	Leaves	Mixed	Vegetation	Exposed Root
Excluded	Invaded	14.23 ± 2.71	0.45 ± 0.2	3.6 ± 1.04	69.45 ± 4.34	12.12 ± 1.96	0	0.19 ± 0.19
Excluded	Removed	10.34 ± 1.42	0	5.63 ± 1.48	77 ± 2.97	7 ± 1.45	0.04 ± 0.04	0
Excluded	Uninvaded	16.45 ± 2.77	0.04 ± 0.04	5.52 ± 1.78	72.78 ± 3.69	4.86 ± 0.9	0.34 ± 0.22	0.04 ± 0.04
Present	Invaded	10.97 ± 2	0.52 ± 0.32	8.34 ± 2.79	76.23 ± 4.17	3.93 ± 1.22	0.04 ± 0.04	0
Present	Removed	13.89 ± 2.69	0.04 ± 0.04	9.04 ± 2.09	64.67 ± 4.39	12.04 ± 2.56	0.19 ± 0.1	0.15 ± 0.15
Present	Uninvaded	29.49 ± 2.76	0	6.15 ± 0.93	58.15 ± 3.63	5.63 ± 1.35	0.6 ± 0.23	0

Appendix 1: ANOVA tables of effects of white-tailed deer and Amur honeysuckle on diversity of invertebrate orders in June, July, August, and September 2005 and 2006.

June 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	9.388889	0.2515	0.6251
honeysuckle	2	24.333333	0.3259	0.7281
deer*honeysuckle	2	2.777778	0.0372	0.9636
Model	5	36.50000	0.1955	0.9583
Error	12	448.00000		
C. Total	17	484.50000		

July 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	2.722222	0.0818	0.7797
honeysuckle	2	75.000000	1.1269	0.3561
deer*honeysuckle	2	7.444444	0.1119	0.8951
Model	5	85.16667	0.5119	0.7625
Error	12	399.33333		
C. Total	17	484.50000		

August 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	112.50000	7.1555	0.0202
honeysuckle	2	181.00000	5.7562	0.0177
deer*honeysuckle	2	2.33333	0.0742	0.9289
Model	5	295.83333	3.7633	0.0279
Error	12	188.66667		
C. Total	17	484.50000		

September 2005

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	6.72222	0.2944	0.5973
honeysuckle	2	186.33333	4.0803	0.0445
deer*honeysuckle	2	15.44444	0.3382	0.7196
Model	5	208.50000	1.8263	0.1821
Error	12	274.00000		
C. Total	17	482.50000		

June 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	12.50000	0.4913	0.4967
honeysuckle	2	156.33333	3.0721	0.0837
deer*honeysuckle	2	10.33333	0.2031	0.8190
Model	5	179.16667	1.4083	0.2894
Error	12	305.33333		
C. Total	17	484.50000		

July 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	24.50000	1.1025	0.3144
honeysuckle	2	177.33333	3.9900	0.0469
deer*honeysuckle	2	16.00000	0.3600	0.7050
Model	5	217.83333	1.9605	0.1575
Error	12	266.66667		
C. Total	17	484.50000		

August 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	20.055556	0.7865	0.3926
honeysuckle	2	76.333333	1.4967	0.2628
deer*honeysuckle	2	82.111111	1.6100	0.2402
Model	5	178.50000	1.4000	0.2921
Error	12	306.00000		
C. Total	17	484.50000		

September 2006

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	20.055556	0.7865	0.3926
honeysuckle	2	76.333333	1.4967	0.2628
deer*honeysuckle	2	82.111111	1.6100	0.2402
Model	5	178.50000	1.4000	0.2921
Error	12	306.00000		
C. Total	17	484.50000		

Appendix 2: ANOVA table of effects of white-tailed deer and Amur honeysuckle on total abundance of litter invertebrates.

Source	DF	Sum of Squares	F Ratio	Prob > F
Month[Year]	6	9.2147237	12.1532	<.0001
Deer	1	1.3196904	10.4431	0.0017
Honey	2	1.0983599	4.3458	0.0156
Deer*Honey	2	0.4789030	1.8949	0.1559
Year	1	0.4959517	3.9246	0.0504
Deer*Year	1	0.0266798	0.2111	0.6469
Honey*Year	2	0.0840643	0.3326	0.7179
Deer*Honey*Year	2	0.5759246	2.2787	0.1079
Month*Deer[Year]	6	0.7925324	1.0453	0.4011
Month*Honey[Year]	12	4.5996874	3.0332	0.0012
Month*Deer*Honey[Year]	12	1.2699627	0.8375	0.6120
Model	47	19.956480	3.3600	<.0001
Error	96	12.131467		
C. Total	143	32.087947		

Appendix 3: ANOVA tables of effects of white-tailed deer and Amur honeysuckle on percent cover of different substrate types.

Bare ground

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.01811207	1.4214	0.2562
honey	2	0.07344472	2.8820	0.0950
deer*honey	2	0.02944406	1.1554	0.3476
Model	5	0.12100086	1.8992	0.1682
Error	12	0.15290563		
C. Total	17	0.27390648		

Woody debris

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.02178779	3.0121	0.1082
honey	2	0.00628576	0.4345	0.6574
deer*honey	2	0.00181812	0.1257	0.8830
Model	5	0.02989167	0.8265	0.5545
Error	12	0.08680164		
C. Total	17	0.11669331		

Leaves

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.03083424	1.1198	0.3108
honey	2	0.01864306	0.3385	0.7194
deer*honey	2	0.04768410	0.8658	0.4454
Model	5	0.09716139	0.7057	0.6302
Error	12	0.33043706		
C. Total	17	0.42759845		

Mixed

Source	DF	Sum of Squares	F Ratio	Prob > F
deer	1	0.00102625	0.1144	0.7410
honey	2	0.02319982	1.2930	0.3101
deer*honey	2	0.04237223	2.3616	0.1365
Model	5	0.06659830	1.4847	0.2656
Error	12	0.10765246		
C. Total	17	0.17425075		

Appendix 4: ANOVA tables of effects of leaf litter depth on diversity and total abundance of invertebrates, and on abundance of the 4 most abundant invertebrate orders.

Diversity

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	0.07256876	3.2590	0.0899
Model	1	0.07256876	3.2590	0.0899
Error	16	0.35627969		
C. Total	17	0.42884844		

Abundance of Araneae

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	0.00001523	0.0005	0.9824
Model	1	0.00001523	0.0005	0.9824
Error	16	0.48712641		
C. Total	17	0.48714165		

Abundance of Acari

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	0.18730709	3.6570	0.0739
Model	1	0.1873071	3.6570	0.0739
Error	16	0.8195087		
C. Total	17	1.0068158		

Abundance of Collembola

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	1.1795907	3.4451	0.0820
Model	1	1.1795907	3.4451	0.0820
Error	16	5.4783141		
C. Total	17	6.6579048		

Abundance of Hymenoptera

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	0.00000785	0.0000	0.9968
Model	1	0.0000078	0.0000	0.9968
Error	16	7.5018640		
C. Total	17	7.5018719		

Total invertebrate abundance

Source	DF	Sum of Squares	F Ratio	Prob > F
depth	1	0.34181445	1.5475	0.2314
Model	1	0.3418144	1.5475	0.2314
Error	16	3.5340197		
C. Total	17	3.8758341		

Appendix 5: ANOVA tables of effects of substrate type on diversity and total abundance of invertebrates, and on abundance of the 4 most abundant invertebrate orders.

Diversity

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.00204897	0.1023	0.7541
woody debris	1	0.00306830	0.1533	0.7018
leaves	1	0.00001135	0.0006	0.9814
mixed	1	0.00062496	0.0312	0.8625
Model	4	0.16858477	2.1052	0.138
Error	13	0.26026367		
C. Total	17	0.42884844		

Abundance of Araneae

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.02583787	0.8780	0.3658
woody debris	1	0.04448402	1.5116	0.2407
leaves	1	0.03418234	1.1615	0.3007
mixed	1	0.02685747	0.9126	0.3569
Model	4	0.10456043	0.8882	0.499
Error	13	0.38258122		
C. Total	17	0.48714165		

Abundance of Acari

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.00419620	0.0741	0.7898
woody debris	1	0.00848685	0.1498	0.7050
leaves	1	0.00034700	0.0061	0.9388
mixed	1	0.00004188	0.0007	0.9787
Model	4	0.2702297	1.1923	0.3600
Error	13	0.7365861		
C. Total	17	1.0068158		

Abundance of Collembola

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.13546328	0.4089	0.5336
woody debris	1	0.00013832	0.0004	0.9840
leaves	1	0.05003965	0.1511	0.7038
mixed	1	0.00566672	0.0171	0.8979
Model	4	2.3516932	1.7749	0.1940
Error	13	4.3062116		
C. Total	17	6.6579048		

Abundance Hymenoptera

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.46413013	1.1763	0.2978
woody debris	1	0.08495677	0.2153	0.6503
leaves	1	0.38288918	0.9704	0.3426
mixed	1	0.24039225	0.6093	0.4490
Model	4	2.3725029	1.5032	0.2582
Error	13	5.1293690		
C. Total	17	7.5018719		

Total invertebrate abundance

Source	DF	Sum of Squares	F Ratio	Prob > F
bare	1	0.18558103	0.9773	0.3409
woody debris	1	0.01691183	0.0891	0.7701
leaves	1	0.11900346	0.6267	0.4428
mixed	1	0.04859438	0.2559	0.6214
Model	4	1.4071242	1.8524	0.1791
Error	13	2.4687099		
C. Total	17	3.8758341		

Chapter 4

Soil and litter effects of invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) in a temperate deciduous forest

Abstract: We examined how invasive Amur honeysuckle (*Lonicera maackii*) and white-tailed deer (*Odocoileus virginianus*) affected decomposition of mixed native leaves and compaction of forest soil. We also quantified effects of Amur honeysuckle on microbial activity in soil contained in greenhouse pots and in soil from an invaded forest. To examine decomposition of leaf litter, we created experimental forest plots representing combinations of deer accessible/excluded and honeysuckle invaded/removed/uninvaded. We then used a combination of field and greenhouse measurements to determine whether honeysuckle altered soil microbial activity. We found that decomposition of leaves of Chinquapin oak (*Quercus muehlenbergii*) and sugar maple (*Acer saccharum*) was similar across deer and honeysuckle treatments. Penetration of forest soil, however, was lower in plots containing either deer or honeysuckle, but removal of honeysuckle and exclusion of deer reduced this effect after two years. We found no interactions, however, of deer and honeysuckle on decomposition of leaf litter or compaction of soil. In our greenhouse experiment, we found that microbial activity was greater in homogenized topsoil when topped with decomposing leaves of honeysuckle than when under leaves of sugar maple. However, we found no difference in microbial activity in soil taken from an invaded area of forest compared to adjacent uninvaded areas. Our results suggest that physical alteration of the soil (e.g., reduced penetrability of soil) was the primary means by which deer or honeysuckle affected forest soil. Although our results suggest that land managers will need to control for both

honeysuckle and deer in order to reduce effects on soil compaction, no active restoration (e.g., tilling, mulching) after control was necessary in our study.

Key words: *Odocoileus virginianus*, *Lonicera maackii*, soil, microbial activity, compaction, litter, decomposition

Introduction

The majority of studies on ecological impacts of invasive plants have focused either on reductions in abundance (Hutchinson and Vankat 1997) and diversity (Dunbar and Facelli 1999) of native plant species, or on alterations in the composition of native plant communities (McKinney 2004). Recently, more emphasis has been placed on ecosystem-level impacts that accompany plant invasions, particularly on soil microbial communities (Vitousek 1990, Kourtev et al. 2002, Duda et al. 2003), litter decomposition (Ashton et al. 2005), and nutrient cycling (Ehrenfeld 2003) and mineralization (Heneghan et al. 2006). If alterations in these attributes of the soil promote growth of invasive species, then impacts of invasive plants on soil function may actually provide a mechanism for their spread into natural systems (Ehrenfeld et al. 2001).

Compared to native species, invasive plants typically have different leaf phenologies and nutrient concentrations, higher growth rates, and novel tissue chemistry, all of which contribute to their impacts on soil (Ehrenfeld 2003, Allison and Vitousek 2004). Such impacts may intensify direct effects of competition between invasive and native plant species by altering fundamental ecosystem functions necessary for native plant survival (Vitousek 1990). Alteration of native plant abundance and diversity, in turn, influences soil microbial communities and rates of litter decomposition (Hector et al. 2000). Allelopathy by some invasive plant species also impacts native species by reducing aboveground biomass or nutrient uptake (Callaway and Aschehoug 2000, Hierro and Callaway 2003). Allelopathic compounds, however, also can alter

composition of soil microbial communities (Batten et al. 2006), and can reduce soil microbial biomass, decomposition, and mineralization of leaf litter (Wardle et al. 1998). Thus, removal of invasive plants may not always be an effective management tool for restoration of native plant communities because true impacts of invasives may not be limited to direct, above ground competition for resources (Heneghan et al. 2006).

Increased decomposition and nutrient availability, particularly nitrogen, was reported under invasive *Tradescantia fluminensis* in a New Zealand lowland forest (Standish et al. 2004), *Sapium sebiferum* (Chinese tallow) in a Texas coastal prairie (Cameron and Spencer 1989), and *Hedychium gardnerianum* (ginger) and *Setaria palmifolia* (bristle grass) in a Hawaiian wet forest (Allison and Vitousek 2004). Alterations in composition of soil microbial communities have been recorded in a serpentine grassland in California invaded by *Centaurea solstitialis* (yellow star thistle) and *Aegilops triuncialis* (barb goatgrass; Batten et al. 2006), and in a deciduous forest in New Jersey invaded by *Microstegium vimineum* (Japanese stilt grass) and *Berberis thunbergii* (Japanese barberry; Kourtev et al. 2002). Such alterations in the structure of soil microbial communities can alter activity of enzymes associated with decomposition and nutrient cycling (Kourtev et al. 2002).

Although invasive plants can have substantial impacts on soil, restoration of habitats invaded by exotic plants is not always straightforward. For example, effects of invasive plants on soil communities may be compounded by over grazing by ungulates who can either increase availability of nitrogen through excretion of urine and feces (Stark et al. 2000), or decrease availability of nitrogen by grazing on plants with high nutrient content, thereby reducing quantity and quality of leaf litter (Hobbs 1996, Tracy and Frank 1998, Tripler et al. 2002). In the later situation, leaf litter from uneaten plants has high C:N ratios and is slow to decompose (Augustine

and McNaughton 1998). This reduces availability of soil nitrogen (Rooney and Waller 2003) and, concomitantly, soil microbial biomass (Sankaran and Augustine 2004). Overabundant ungulates also increase soil compaction, alter soil microclimate, and reduce water infiltration into soil (Yates et al. 2000).

Amur honeysuckle (*Lonicera maackii*; hereafter, honeysuckle) is a fast-growing, invasive shrub introduced to North American from Asia in 1896 for use in horticulture (Luken and Thieret 1995). In invaded regions, Amur honeysuckle reduces abundance (Hutchinson and Vankat 1997) and diversity (Collier et al. 2002) of native herbs, and survival (Gorchov and Trisel 2003, Hartman and McCarthy 2004), density, and species richness of native tree seedlings. (Hutchinson and Vankat 1997, Collier et al. 2002). These impacts on diversity and abundance of native plant species may alter soil microbial communities and litter decomposition (Hector et al. 2000), indirectly contributing to impacts on the plant community (Hooper and Vitousek 1997, Ehrenfeld and Scott 2001). Furthermore, extracts of honeysuckle leaves were dominated by two flavones, apigenin and luteolin, which inhibited germination of *Arabidopsis thaliana* and deterred feeding by the generalist herbivore *Spodoptera exigua* (beet armyworm; Cipollini et al. 2008). In addition to deterring herbivores, some allelocompounds also can reduce microbial activity in soil, lowering rates of litter decomposition (Bassman 2004). However, both apigenin and luteolin have been associated with increased bacterial growth in the rhizospheres of plants exuding these compounds (Phillips and Tsai 1992). Whether this is true in soil invaded by honeysuckle, however, is unknown.

Grazing by over-abundant white-tailed deer (*Odocoileus virginianus*; hereafter, deer) could exacerbate effects of honeysuckle. Deer reduced available nitrogen in soil and leaf tissue in a Minnesota savannah by reducing cover of legumous plant species (Ritchie et al. 1998).

Alternatively, deer can have positive effects on some N-fixing legumes, and, consequently, on soil nitrogen (Bowers and Sacchi 1991). Overabundant deer also reduce abundance, diversity, and composition of native plants (Rooney and Waller 2003, Russell et al 2001), indirectly contributing to alterations in the soil microbial community (Hector et al. 2000). Previous studies have quantified effects of large ungulates such as livestock on soil compaction (Stephenson and Veigel 1987). However, given their abundance, there is a surprising lack of experimental studies to determine whether deer reduce the penetrability (i.e., increase compaction) of forest soil, which would also reduce soil habitat space for detritivores and alter turnover rates of microbial communities (Van Veen and Kiukman 1990), and reduce litter decomposition. Indirectly, reduced penetrability of soil alters structure of plant communities (Habek 1960), which can indirectly reduce decomposition and nutrient cycling (Melillo et al. 1982, Hector et al. 2000).

Because honeysuckle could potentially impact activity of soil microbes, and because both honeysuckle and deer can alter litter decomposition and penetrability of soil, we sought to determine whether invasion by honeysuckle and/or presence of deer altered decomposition of native leaf litter. We also hypothesized that penetration of forest soil would be lower in areas accessible to deer than in areas from which deer were excluded, and higher in areas invaded by honeysuckle than in areas with no history of honeysuckle invasion. Further, we hypothesized that Amur honeysuckle would increase activity of soil microbes.

Methods

Litter Decomposition and Compaction of Soil

We studied a second-growth hardwood forest at the Cincinnati Nature Center (CNC), a 405-hectare nature preserve in Milford, Ohio dominated by beech (*Fagus grandifolia*), Chinquapin oak (*Quercus muehlenbergii*), red oak (*Quercus rubra*), shagbark hickory (*Carya ovata*),

bitternut hickory (*Carya cordiformis*), red maple (*Acer rubrum*), and sugar maple (*Acer saccharum*). Our research site was located on a west-facing slope bisected by a gully (2 m wide, 6 m deep) running from east to west. In April 2005, we positioned twelve (10 x 10-m) plots in an area north of the gully that was heavily invaded by honeysuckle, and six plots in an area south of the gully that had no history of invasion by honeysuckle. Treatments were randomly assigned to plots in a 3 x 2 factorial design representing *L. maackii* present/absent/removed and *O. virginianus* present/absent. There were three replicates per treatment type. We removed all honeysuckle shrubs from half of our invaded plots by cutting them at the base and saturating the cut stem with 2% glyphosate. Commercial deer fencing (Benner's Gardens, Conshohocken, PA, www.bennersgardens.com) was placed around six invaded and three uninvaded plots to exclude deer.

In late September 2005, we collected freshly fallen leaves from Chinquapin oak (*Quercus muehlenbergii*) and sugar maple (*Acer saccharum*) from our study area, and dried them at room temperature for one month. Once dried, approximately one gram of whole leaves of each species (two grams total per bag) was placed into 20 x 20-cm nylon litter bags constructed of 3-mm mesh on the side facing the ground and 15-mm mesh on the top. Each bag was individually labeled with a metal tag. Twenty-five bags were pinned to the ground under the leaf litter randomly in each plot on 7 April 2006. From May through October 2006, three bags were collected each month from each plot, transported to the lab in individual plastic bags, individually rinsed in fresh standing water to remove dust and caked mud, and dried at 60° C for 48 hours. Dried leaf material was then sorted from fine roots and other debris, and weighed. Bags remaining in experimental plots were collected in late March 2007 to compare over-winter mass loss in each treatment. Reduction in litter mass was used to quantify litter decomposition.

Soil penetrability was estimated in June 2006 using a static Lang™ penetrometer that measured soil resistance to penetration along an index range from one to 20. In each plot, we measured penetration in ten arbitrary locations, but avoided exposed rock and woody debris. Index values were compared to a standard curve to obtain force (kg) required to penetrate the soil (Lang Penetrometer, Inc., Gulf Shores, AL, USA).

Microbial Activity

Greenhouse Methods

Leaves from honeysuckle shrubs were collected before they abscised in October 2005 from areas near the University of Cincinnati campus. Leaves were dried at ambient temperature and frozen. Soil was collected in January 2006 from the A-horizon (<10 cm depth) in an area at CNC that had no history of invasion by honeysuckle. We used left-over sugar maple leaves remaining from litter decomposition bags as a native control. Soil was brought to a greenhouse at the University of Cincinnati and sifted through 5-mm poultry wire. All visible organic matter, including fine roots and leaf debris, was removed with forceps. In February 2006, We placed 350 grams of homogenized topsoil into each of twenty 15-cm plastic pots and added tap water to bring the soil to field capacity. Samples were then left undisturbed for one week to control for artificially increased microbial activity due to mixing the soil (Öhlinger 1996). Then, 5g of whole leaves of honeysuckle were placed on top of soil in ten of these pots, and 5g of whole leaves of sugar maple were placed on the soil surface in the remaining ten pots. All leaves had first been dried at room temperature, and reconstituted with distilled water to control for differences in surface bacteria or fungi that could have affected measurements of decomposition and microbial activity. Three 2g samples of soil were taken from the upper 1cm of soil in each pot and placed into 25-

ml test tubes every two weeks for 20 weeks. These samples were then analyzed for soil microbial activity using methods described below.

Throughout this experiment, pots were exposed to full light, but soil was kept at field capacity, and pots were randomly arranged and regularly rotated. The greenhouse was maintained at 25.3° C (77.5° F).

Field Methods

We chose to measure soil microbial activity in the field before (July) and after (October) canopy tree leaves abscised in 2006. In each of these months, we selected nine forested areas within Woodland Mound, a 398-hectare second-growth forest preserve in Cincinnati, Ohio similar to CNC. All areas were approximately 25-m apart, but varied in area. Three areas were invaded by honeysuckle, three were uninvaded, and three had been sprayed with 2% glyphosate to kill honeysuckle before native plants leafed out in early spring in 2006. In the middle of each selected area, we collected five 500-g soil samples from the top 2-cm of topsoil. Soil samples were returned to lab, sifted through a 5-mm sieve, and all fine organic debris was removed. These samples also were then left undisturbed for one week to control for artificially increased microbial activity due to mixing the soil (Öhlinger 1996). To determine differences in microbial activity, two grams of soil from each sample (n = 15 per area) were analyzed for soil microbial activity.

Measurement of Soil Microbial Activity

Soil microbial activity in greenhouse and field samples was quantified by measuring dehydrogenase activity of soil microbes using the triphenyltetrazolium chloride reduction (TTC) method. This method relies on the reduction of TTC to triphenylformazan (TPF) (Casida et al. 1964). More TTC is reduced to TPF at higher microbial activity, and final concentrations of TPF

can be used to estimate soil microbial activity. TPF is a red pigment; at 485 nm, absorbance of TPF is equivalent to its concentration (Casida et al. 1964, Friedel et al. 1994).

To measure soil microbial activity, 2g soil samples were mixed with 2 ml 0.5% buffered (7.6 pH) TTC solution. The soil-TTC mixture was then incubated at 30° C for 24 hours. After incubation, TPF was extracted by adding 20 ml acetone to each sample and placing it into a shaking incubator (200 rpm) at 30° C for 2 hours. The supernatant from each tube was suction-filtered through filter paper (1.5 µm). The soil remaining in each tube was flushed twice with 5 ml acetone, and these supernatants filtered. Absorbance of the final TPF filtrate for each tube was measured with a spectrophotometer (absorbance = 485 nm). To control for slight changes in TPF concentration during filtration, a standard curve of TPF concentration versus absorbance was created by filtering known concentrations of TPF using the same protocol (Casida et al. 1964, Friedel et al. 1994, Ohlinger 1996, Sigler and Zeyer 2002).

Statistical Analyses

Percent litter mass remaining in each litter bag was analyzed by two-way repeated measures ANOVA (JMP IN, version 5.1.2; SAS Institute Inc., Cary, NC) to determine differences among honeysuckle and deer treatments. An angular transformation was used to meet assumptions of repeated measures ANOVA. Soil penetration measurements were averaged for each plot, transformed by exp(x) to attain normality, and compared among treatments with two-way ANOVA.

Differences in microbial activity between pots with honeysuckle or sugar maple were determined by calculating mean µg TPF of the 3 sub-samples from each pot. We then compared differences in mean µg TPF using repeated measures ANOVA on log-transformed data. For field-collected soil, we calculated mean µg TPF for each of the 9 forested areas in Woodland

Mound, resulting in 3 replicates per honeysuckle habitat type (i.e., invaded, uninvaded, killed). Data met all assumptions of ANOVA, and differences in mean µg TPF were compared with two-way ANOVA on untransformed data. When applicable, Tukey HSD post-hoc tests ($\alpha = 0.05$) were used to test pair-wise differences between honeysuckle treatments.

Results

Decomposition: Neither honeysuckle ($F_{2,48} = 0.95$, $p = 0.39$) nor deer ($F_{1,48} = 0.79$, $p = 0.38$) altered decomposition of leaf litter (Fig. 1). There also was no interaction of deer and honeysuckle on decomposition ($F_{2,48} = 0.33$, $p = 0.72$). In all plots, leaf litter in over-wintered bags had completely decayed by the end of March 2007.

Soil Penetration: Soil penetration was lower in plots accessible to deer ($F_{1,12} = 10.57$, $p = 0.01$) than in plots from which deer were excluded. Honeysuckle treatment type also affected penetration of soil ($F_{2,12} = 13.13$, $p = 0.001$; Table 1); soil in plots containing honeysuckle had lower penetration than soil in plots from which honeysuckle had been removed or had never invaded There was no significant deer x honeysuckle interaction on soil penetrability ($F_{2,12} = 3.15$, $p = 0.08$)

Greenhouse: Microbial activity in soil beneath honeysuckle leaves was higher than in soil beneath sugar maple ($F_{1,58} = 22.74$, $p < 0.0001$; Fig. 2). There also was a significant effect of time ($F_{9,50} = 68.9$, $p < 0.0001$) on microbial activity, and a significant time x leaf type interaction ($F_{9,50} = 3.05$, $p < 0.006$). Leaves of honeysuckle had completely decomposed by week 8, but leaves of sugar maple had not completely decayed when pots were removed from the greenhouse at week 40. We analyzed the first 8 weeks when honeysuckle was decomposing and found that microbial activity was significantly greater in pots with honeysuckle than pots with sugar maple

($F_{3,16} = 4.47$, $p = 0.018$). However, there was no difference in microbial activity between pots with honeysuckle or sugar maple in weeks 9 through 20 ($F_{1,18} = 2.21$, $p = 0.15$; Fig. 2).

Field: Microbial activity did not differ between July and October ($F_{1,12} = 0.01$, $p = 0.94$), and there was no habitat x month interaction ($F_{2,12} = 1.05$, $p = 0.38$), indicating effects were similar in both months (Fig. 3). Soil microbial activity was greater in invaded areas where sugar maple had been treated with glyphosate than in invaded areas that had not been treated ($F_{2,2} = 6.58$, $p < 0.01$; Appendix 1).

Discussion

Our hypothesis that soil penetrability in plots accessible to deer would be lower compared to plots from which deer were excluded was support. However, penetrability of soil was significantly higher in deer exclusion plots after two years of deer exclusion. By comparison, reduced penetrability from grazing by livestock can take four years to recover (Stephenson and Veigel 1987). By altering soil structure, deer can directly affect soil microbial communities by reducing interstitial spaces that serve as habitat for soil organisms (Van Veen and Kiukman 1990). Compacted soil absorbs less water and over time becomes anaerobic, changes that reduce root growth and above ground plant biomass (Whalley et al. 1995). These indirect effects of grazing by deer intensify direct impacts of grazing (Rooney and Waller 2003). However, despite the impact of deer on soil penetrability, there was no effect of deer on decomposition of native leaf litter, indicating that reduced penetrability of soil did not alter soil function.

Lower penetration of soil in plots containing honeysuckle also supported our hypothesis. This reduced penetrability in invaded plots can be attributed to their thick, shallow, and matted root systems. However, soil penetrability was similar in uninvaded and honeysuckle removal

plots two years after removal of honeysuckle crowns. When we removed crowns of honeysuckle, root systems were left intact. Increased penetrability of soil in honeysuckle removal plots, therefore, suggests that roots decomposed rapidly. While invasion of honeysuckle was accompanied by reduced penetrability of the soil, removal of the shrub crown reversed this impact. As with deer, however, reduced penetrability of soil in invaded plots was not accompanied by functional impacts; litter decomposition was similar among honeysuckle treatments.

Leaves of honeysuckle decomposed more than five times faster than leaves of sugar maple in greenhouse pots, and microbial activity in soil under honeysuckle leaves was greater than in soil under sugar maple leaves, refuting our hypothesis. Once honeysuckle leaves had fully decayed, soil microbes no longer had substrate on which to feed, and microbial activity diminished, but still was equivalent to microbial activity in sugar maple pots. Soil microbial activity may have been stimulated by the release of allelopathic compounds (e.g., apigenin and luteolin) during decomposition of honeysuckle (Phillips and Tsai 1992, Cipollini et al. 2008). Although apigenin and luteolin have been extracted from leaves of honeysuckle (Cipollini et al. 2008), release of these compounds during decomposition of honeysuckle in the field has not been established. While no apigenin compounds have been found in extracts of sugar maple leaves, a luteolin derivative has been identified, but was not a major constituent of sugar maple leaves (Delendick 1990).

Decomposition rates of leaves are typically a function of foliar lignin:nitrogen ratios, rather than simply a response to concentration of foliar nitrogen (Melillo et al. 1982). Differences in decomposition and subsequent higher activity of soil microbes in greenhouse pots, therefore, could reflect differences in lignin:nitrogen ratios between honeysuckle and sugar maple leaves

(Melillo et al. 1982). Foliar nitrogen content of honeysuckle was 1.82 % ± 0.049 in forest interiors (Lieurance 2004), while nitrogen content of sugar maple ranged from 0.6 to 0.83% (Mellilo et al, 1982, McClaughtery et al 1985). Lignin concentration of sugar maple leaves was reported as 10.1% (Melillo et al. 1982), but lignin content of honeysuckle has not been quantified. However, our sugar maple leaves were collected in autumn, after resorption of foliar nitrogen (Ryan and Borman 1982). Honeysuckle, however, becomes exposed to cold temperatures before its leaves senesce, which likely impairs nutrient resorption (Demars and Boerner 1997). Thus, the differences in age of leaves and timing of leaf collection could have led to differences in foliar nitrogen, and subsequently to differences in soil microbial activity.

Given our results from greenhouse pots, we expected that microbial activity would also be higher in soil under honeysuckle in the field. However, soil microbial activity in invaded areas was similar to that in uninvaded areas. One explanation for this could be related to our sampling scheme. Leaves of honeysuckle abscise in November (Trisel 1997) after native species have already fallen, but we sampled in June and October. Hence, decomposition of honeysuckle leaves that had fallen the previous year would have been complete, and any effects on soil microbes would have dissipated before we sampled. In the field, there was never a layer of decomposing honeysuckle leaves as there was on greenhouse pots, so our forest soil samples were not strictly comparable to those taken from beneath honeysuckle leaves in the greenhouse pots.

Although soil microbial activity was similar in invaded and uninvaded areas at Woodland Mound, soil from areas where honeysuckle had been killed with glyphosate had higher microbial activity than soil from areas where shrubs were untreated. When applied to soil, glyphosate increases microbial enzyme activity because it is a source of available carbon and nitrogen

(Haney et al. 2000). While glyphosate is quickly metabolized in soil, residual effects on microbial activity can last up to 56 days after application (Haney et al. 2000). In our case, stands of honeysuckle had been treated with glyphosate at least three months before our July soil samples were taken. Therefore, increased microbial activity in glyphosate-treated areas was unlikely to be a direct result of glyphosate treatment, particularly in October. Instead, an indirect effect of treatment may be more plausible. In areas treated with glyphosate, abundant dead woody debris, including small branches of dead honeysuckle could support increased biomass of both saprophytic and ectomycorrhizal fungi (Tedersoo et al. 2003, Buée et al. 2007). Decomposing roots of killed honeysuckle could also have contributed to this effect. Because TTC reduction does not differentiate between bacterial and fungal contributions to microbial activity, increased microbial activity in treated areas could have been due to greater fungal biomass in these areas.

Our results demonstrated that both deer and honeysuckle penetrability of forest soil. However, honeysuckle did not reduce soil microbial activity, and neither honeysuckle nor deer altered decomposition of native leaves. This is in contrast to previous studies that have shown increased decomposition in areas invaded by exotic plants (Cameron and Spencer 1989, Allison and Vitousek 2004, Standish et al. 2004). Decomposing leaves of honeysuckle sustained higher soil microbial activity than leaves of sugar maple in greenhouse pots, but field samples provided inconclusive data as to whether honeysuckle alters soil microbial activity in invaded forests.

Our findings are encouraging for land managers in the Midwest. In our study, physical alteration of soil structure through reduced penetrability of soil appeared to be the principal means by which deer and honeysuckle affect forest soil, but this effect was reduced in 2 years after removal of honeysuckle and exclusion of deer. Although we found no interactions of deer

and honeysuckle on penetrability of soil, the significant decrease in soil penetrability in the presence of either deer or honeysuckle indicates that managers will need to control both species to improve soil quality. However, our data also indicate that removal of honeysuckle crowns and exclusion or culling of deer should increase penetrability of soil without additional active restoration (e.g., tilling, mulching) after removal or exclusion.

We suggest that future studies focus on whether soil microbial activity is affected in November, when honeysuckle leaves abscise and begin to decompose. It also would be valuable to determine whether honeysuckle alters the composition or biomass of the microbial community of the soil, and whether these changes improve growth or germination of honeysuckle, providing another mechanism where by invasiveness of honeysuckle is increased. Lastly, effects of overabundant deer on microbial communities of forest soil have not been examined, but these data may reveal indirect effects of deer on native plant communities.

Acknowledgments

We thank T. Culley, D. Gorchov, E. Maurer, and G. Uetz for exceptional advice and willingness to assist with our experimental design, data analysis, and interpretation of the results. We are also appreciative of B. Kinkle and J. Shann for helping with methodology and data interpretation. Lastly, we thank A. Cooperman, M. Cramer, E. Dame, G. Klein, J. Lawrence, and S. Puthoff for providing invaluable assistance in the lab and field.

Literature Cited

Allison, S.D. and P.M. Vitousek. 2004. Rapid nutrient cycling in leaf litter from invasive plants in Hawai'i. Oecologia 141:612-619.

Ashton, I.W., L.A. Hyatt, K.M. Howe, J. Gurevitch, and M.T. Lerdau. 2005. Invasive species accelerate decomposition and litter nitrogen loss in a mixed deciduous forest. Ecological Applications 15:1263-1272.

Augustine, D.J. and S.J. McNaughton. 1998. Ungulate effects on the functional species composition of plant communities: herbivore selectivity and plant tolerance. The Journal of Wildlife Management 62:1165-1183.

Bassman, J.H. 2004. Ecosystem consequence of enhanced solar ultraviolet radiation: secondary plant metabolites as mediators of multiple trophic interactions in terrestrial plant communities. Photochemistry and Photobiology 79: 382-398.

Batten, K.M., K.M. Snow, K.F. Davies, and S.P. Harrison. 2006. Two invasive plants alter soil microbial community composition in serpentine grasslands. Biological Invasions 8:217-230.

Boisvert, J.M., C. Cloutier, and J. McNeil. 1981. *Hydaphis tartaricae* (homoptera: Aphididae), a pest of honeysuckle new to North America. The Canadian Entomologist 113:415-418.

Bowers, M.A. and C.F. Sacchi. 1991. Fungal mediation of a plant-herbivore interaction in an early successional plant community. 72: 1032-1037.

Buée, M., P.E. Courty, D. Mignot, and J. Garbaye. 2007. Soil niche effect on species diversity and catabolic activities in an ectomycorrhizal fungal community. Soil Biology and Biochemistry 39:1947-1955.

Callaway, R.M. and E.T. Aschehoug. 2000. Invasive plants versus their new and old neighbors: a mechanism for exotic invasion. Science 290:521-523.

Cameron, G.N., and S.R. Spencer. 1989. Rapid leaf decay and nutrient release in a Chinese tallow forest. Oecologia 80: 222-228.

Casida, L.E., D.A. Klein, and T. Santoro. 1964. Soil dehydrogenase activity. Soil Science 98:371-376.

Cipollini, D., R. Stevenson, S. Enright, A. Eyles, and P. Bonello. 2008. Phenolic metabolites in leaves of the invasive shrub, *Lonicera maackii*, and their potential phytotoxic and anti-herbivory effects. Journal of Chemical Ecology 34:144-152.

Collier, M.H. and J.L. Vankat. 2002. Diminished plant richness and abundance below *Lonicera maackii*, and invasive shrub. The American Midland Naturalist 147:60-71.

Delendick, T. J. 1990. A survey of foliar flavonoids in the Aceraceae. Memoirs of the New York Botanical Gardens no. 54. New York Botanical Garden Press. New York.

Demars, B.G. and R.E.J. Boerner. 1997. Foliar nutrient dynamics and resorption in naturalized *Loniera maackii* (Caprifoliaceae) populations in Ohio, USA. American Journal of Botany 84:112-117.

Dorning, M. and D. Cipollini. 2006. Leaf and root extracts of the invasive shrub, *Lonicera maackii*, inhibit seed germination of three herbs with no autotoxic effects. Plant Ecology 184:287-296.

Duda, J.J., D.C. Freeman, J.M. Emlen, J. Belnap, S.G. Kitchen, J.C. Zak, E. Sobek, M. Tracy, and J. Montante. 2003. Differences in native soil ecology associated with invasion of the exotic annual chenopod, *Halogeton glomeratus*.

Dunbar, K.R. and J.M. Facelli. 1999. The impact of a novel invasive species, *Orbea variegata* (African carrion flower), on the chenopod shrublands of South Australia. Journal of Arid Environments 41: 37-48.

Ehrenfeld, J.G., P. Kourtev, and W. Huang. 2001. Changes in soil functions following invasions of exotic understory plants in deciduous forests. Ecological Applications 11:1287-1300.

Ehrenfeld, J.G. and N. Scott. 2001. Invasive species and the soil: Effects on organisms and ecosystem processes. Ecological Applications 11:1259-1260.

Ehrenfeld, J.G. 2003. Effects of exotic plant invasions on soil nutrient cycling processes. Ecosystems 6:503-523.

Friedel, J.K., K. Mölter, and W.R. Fischer. 1994. Comparison and improvement of methods for determining soil dehydrogenase activity by using triphenyltetrzolium chloride and iodonitrotetrzolium chloride. Biology and Fertility of Soil 18:291-296.

Gorchov, D.L. and D.E. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology 166: 13-24.

Habek, J.R. 1960. Winter deer activity in the white cedar swamps of northern Wisconsin. Ecology 41:327-333.

Haney, R.L., S.A. Senseman, F.M. Hons, and D.A. Zuberer. 2000. Effect of glyphosate on soil microbial activity and biomass. Weed Science 48: 89-93.

Hartman, K.M. and B.C. McCarthy. 2004. Restoration of a forest understory after the removal of an invasive shrub, Amur honeysuckle (*Lonicera maackii*). Restoration Ecology 12:154-165.

Hector, A., A.J. Beale, A. Minns, S.J. Otway, and J.H. Lawton. 2000. Consequences of the reduction of plant diversity for litter decomposition: effects through litter quality and microenvironment. Oikos 90: 357-371.

Heneghan, L., F. Fatemi, L. Umek, K. Grady, K. Fagen, and M. Workman. The invasive shrub European buckthorn (*Rhamnus cathartica*, L.) alters soil properties in Midwestern U.S. woodlands. Applied Soil Ecology 32: 142-148.

Herman, D.E. and C.G. Davidson. 1997. Evaluation of *Lonicera* taxa for honeysuckle aphid susceptibility, winter hardiness and use. Journal of Environmental Horticulture 15:177-182.

Hierro, J.L. and R.M. Callaway. 2003. Allelopathy and exotic plant invasion. Plant and Soil 256:29-39.

Hobbs, N.T. 1996. Modification of ecosystems by ungulates. The Journal of Wildlife Management 60:695-713.

Hooper, D.U. and P.M. Vitousek. 1997. The effects of plant composition and diversity on ecosystem processes. Science 277:1302-1305.

Hunter, M.D., S. Adl, C.M. Pringle, and D.C. Coleman. 2003. Relative effects of macroinvertebrates and habitat on the chemistry of litter during decomposition. Pedobiologia 47:101-115.

Hutchinson, T.F. and J.L. Vankat. 1997. Invasibility and effects of *Lonicera maackii* in southwestern Ohio forests. Conservation Biology 11: 1117-1124.

Kourtev, P.S., J.G. Ehrenfeld, and M. Häggblom. 2002. Exotic plant species alter the microbial community structure and unction in the soil. Ecology 83:3152-3166.

Lieurance, D.M. 2004. Leaf phenology, fecundity, and biomass allocation of the invasive shrubs *Lonicera maackii* (Rupr.) Maxim in the contrasting light environments. M.S. Thesis, Ohio University, Athens, Ohio.

Luken, J.O. and J.W. Thieret. 1995 Amur honeysuckle (*Lonicera maackii*; Caprifoliaceae): its ascent, decline, and fall. SIDA 16:479-503.

McClaughtery, C.A., J. Pastor, J.D. Aber, and J.M. Melillo. Forest litter decomposition in relation to soil nitrogen dynamics and litter quality. Ecology 66:266-275.

McKinney, M.L. 2004. Measuring floristic homogenization by non-native plants in North America. Global Ecology and Biogeography 13:47-53.

Melillo, J.M., J.D. Aber, and J.F. Muratore. 1982. Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. Ecology 63: 621-626.

Öhlinger R..1996. Dehydrogenase activity with the substrate TTC. *In*: Methods in Soil Biology. Schinner F., R. Öhlinger, E. Kandeler, and R. Margesin (eds). Springer Verlag. Berlin.

Phillips, D.A. and S.M. Tsai. 1992. Flavonoids as plant signals to rhizosphere microbes. Mycorrhiza 1:55-58.

Ritchie, M.E., D. Tilman, and J.M.H. Knops. 1998. Herbivore effects on plant and nitrogen dynamics in oak savanna. Ecology 79:165-177.

Rooney, T.P., and D.M. Waller. 2003. Direct and indirect effects of white-tailed deer in forest ecosystems. Forest Ecology and Management 181:165-176.

Russell, F.L., D.B. Zippin, and N.L. Fowler. 2001. Effects of white-tailed deer (*Odocoileus virginianus*) on plants, plant populations and communities: a review. The American Midland Naturalist 146:1-26.

Ryan, D.F. and F.H. Bormann. 1982. Nutrient resorption in northern hardwood forests. Bioscience 32:29-32.

Sankaran, M. and D.J. Augustine. 2004. Large herbivores suppress decomposer abundance in a semiarid grazing ecosystem. Ecology 85:1052-1061.

Sigler, W.V. and J. Zeyer. 2002. Microbial diversity and activity along the forefields of two receding glaciers. Microbial Ecology 43:397-407.

Standish, R.J., P.A. Williams, A.W. Robertson, N.A. Scott, and D.I. Hedderley. 2004. Invasion by a perennial herb increases decomposition rate and alters nutrient availability in warm temperate lowland forest remnants. Biological Invasions 6: 71-81.

Stark, S., D.A. Wardle, R. Ohtonen, T. Helle, and G.W. Yeates. 2000. The effect of reindeer grazing on decomposition, mineralization and soil biota in a dry oligotrophic Scots pine forest. Oikos 90:301-310.

Stephenson, G.R. and A. Viegel. Recovery of compacted soil on pastures used for winter cattle feeding. Journal of Range Management 40:46-48.

Tedersoo, L., U. Kõljalg, N. Hallenberg, and K.H. Larsson. 2003. Fine scale distribution of ectomycorrhizal fungi and roots across substrate layers including coarse woody debris in a mixed forest. New Phytologist 159:153-165.

Tracy, B.F. and D.A. Frank. 1998. Herbivore influence on soil microbial biomass and nitrogen mineralization in a northern grassland ecosystem: Yellowstone National Park. Oecologia 114:556-562.

Tripler, C.E., C.D. Canham, R.S. Inouye, and J.L. Schnurr. 2002. Soil nitrogen availability, plant luxury consumption, and herbivory by white-tailed deer. Oecologia 133: 517-524.

Trisel, D.E. 1997. The invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae): factors contributing to its success and its effect on native species. Ph.D. Dissertation, Miami University, Oxford, Ohio.

Van Veen, J.A. and P.J. Kuikman. 1990. Soil structural aspects of decomposition of organic matter by micro-organisms. Biogeochemistry 11:213-233.

Verhoef, H.A. and L. Brussard. 1990. Decomposition and nitrogen mineralization in natural and agro-ecosystems: the contribution of soil animals. Biogeochemistry: 11:175-211.

Vitousek, P.M. 1990. Biological Invasions and ecosystem processes: towards an integration of population biology and ecosystem studies. Oikos 57:7-13.

Wardle, D.A., M.C. Nilsson, C. Gallett, and O. Zackrisson. 1998. An ecosystem-level perspective of allelopathy. Biological reviews of the Cambridge Philosophical Society 73:305-319.

Yates, C.J., D.A. Norton, and R.J. Hobbs. 2000. Grazing effects on plant cover, soil and microclimate in fragmented woodlands in south-western Australia: implications for restoration. Austral Ecology 25:36-47.

Figure 1: Comparison of leaf litter decomposition among treatments. Mean percent litter mass remaining in litter decomposition bags in each treatment type is shown (± S.E.).

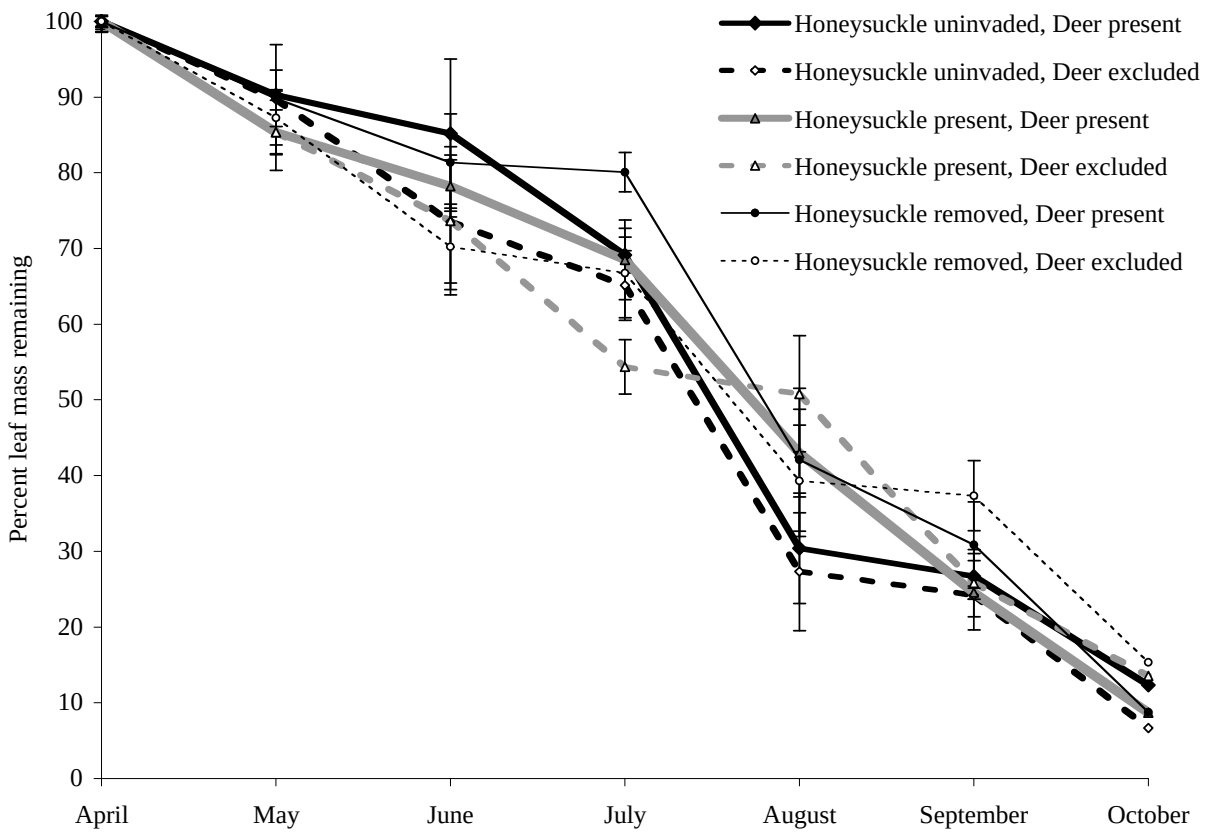

Percent leaf mass remaining
100
90
80
70
60
50
40
30
20
10
0
April
May
June
July
August
September
October
Honeysuckle uninvaded, Deer present
Honeysuckle uninvaded, Deer excluded
Honeysuckle present, Deer present
Honeysuckle present, Deer excluded
Honeysuckle removed, Deer present
Honeysuckle removed, Deer excluded

Figure 2: Comparison of microbial activity in soil from greenhouse pots containing either honeysuckle or sugar maple, as measured by reduction of TTC to TPF (mean µg / g TTC / 16 hr ± S.E.). Dashed line represents point at which all honeysuckle litter had decomposed.

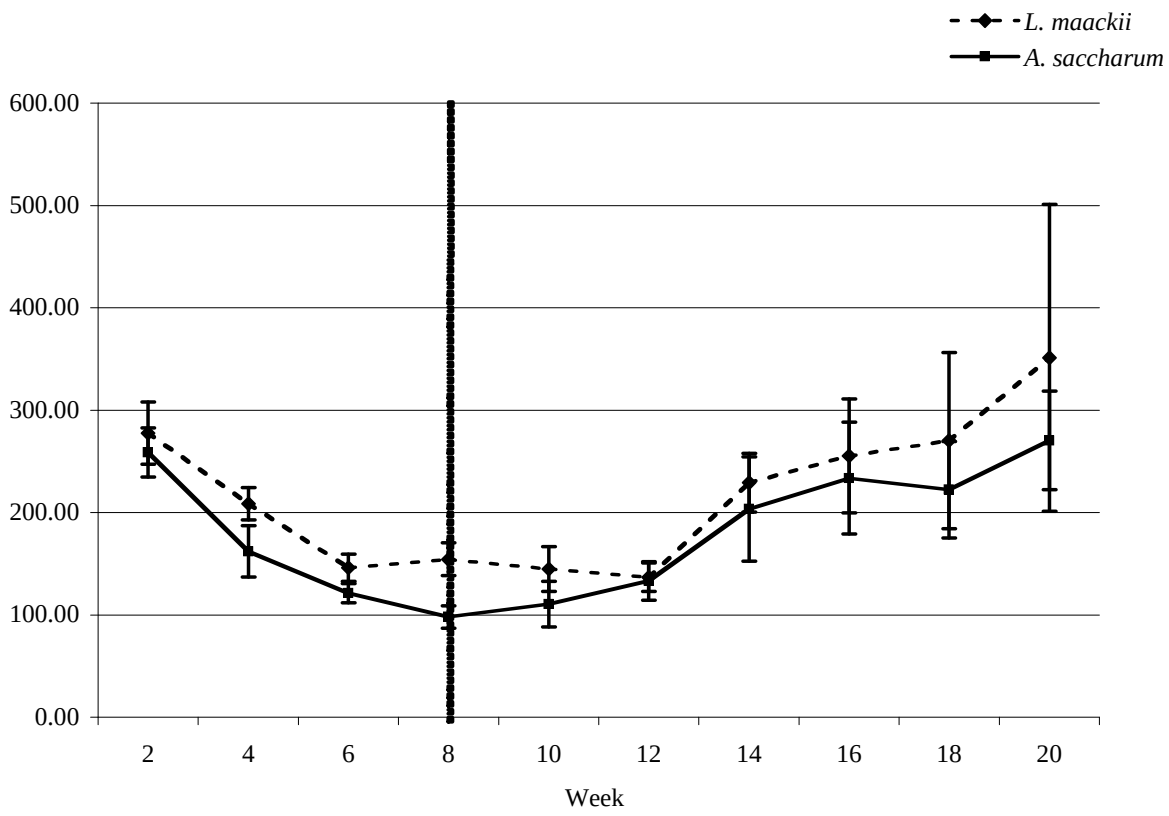

L. maackii
A. saccharum
600.00
500.00
400.00
300.00
200.00
100.00
0.00
2
4
6
8
10
12
14
16
18
20
Week

Figure 3: Comparison of microbial activity in soil from different honeysuckle treatments in forested plots at Woodland Mound Park in July (white columns) and October (black columns), as measured by reduction of TTC to TPF (mean µg / g TTC / 16 hr ± S.E.).

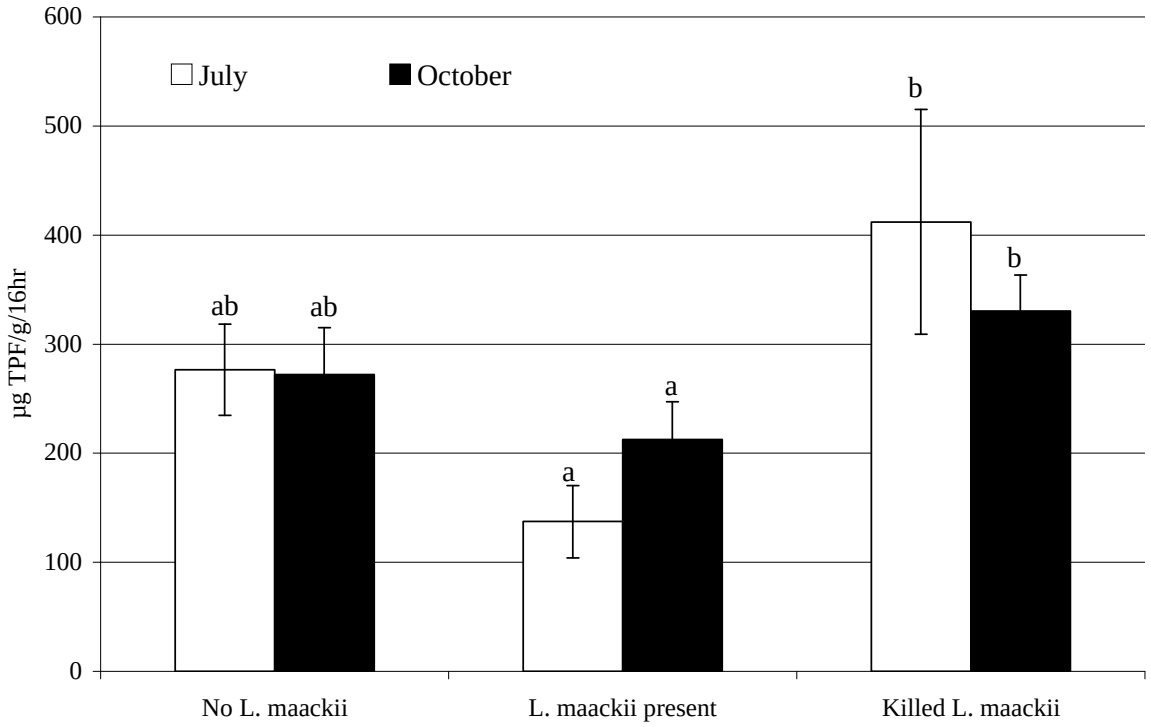

600
500
400
300
200
100
0
µg TPF/g/16hr
July
October
ab
ab
a
a
b
b
No L. maackii
L. maackii present
Killed L. maackii

Table 1: Soil compaction, as estimated by mean (± SE) kg force required to penetrate soil

Honeysuckle	Deer	Force required (kg)
Uninvaded	Excluded	5.90 ± 0.41
Uninvaded	Present	7.13 ± 0.17
Invaded	Excluded	7.20 ± 0.14
Invaded	Present	7.37 ± 0.10
Removed	Excluded	6.51 ± 0.05
Removed	Present	6.68 ± 0.13

Appendix 1: ANOVA comparison of soil microbial activity across honeysuckle treatments (invaded, uninvaded, killed) in forested areas of Woodland Mound Park.

Source	DF	Sum of Squares	F Ratio	Prob > F
honeysuckle	2	115606.21	6.5826	0.0118
month	1	57.96	0.0066	0.9366
honeysuckle*month	2	18441.04	1.0500	0.3800
Model	5	134105.21	3.0544	0.0525
Error	12	105373.72		
C. Total	17	239478.93		

Chapter Five: Conclusions

Amur honeysuckle and white-tailed deer both significantly affected diversity, abundance, and community composition of forest herbs and litter-dwelling invertebrates, but these effects were temporally, variable and were particularly inconsistent for Amur honeysuckle. We also found no effect of type of litter substrate or litter depth on diversity or abundance of litter invertebrates. These results indicated that alteration of the native plant community and structure of the litter layer by honeysuckle and deer was either not great enough to elicit responses from litter-dwelling invertebrates or that litter invertebrates were more influenced by habitat characteristics that were not measured, such as vegetative cover or litter moisture or temperature.

There were few significant interactions of Amur honeysuckle or white-tailed deer on native plant and litter-dwelling invertebrate communities, litter decomposition, or compaction of soil. However, the finding that honeysuckle appeared to protect false Solomon's seal from grazing by deer has important management implications. A similar protective function of honeysuckle was shown for red oak (*Quercus rubra*) seedlings (Gorchov and Trisel 2003). If such a protective function by Amur honeysuckle is widespread, then removal of Amur honeysuckle from areas impacted by both Amur honeysuckle and white-tailed deer could prove ineffective at restoring native plant communities if not also accompanied by management of white-tailed deer. Before implementing large-scale removal of Amur honeysuckle, land managers should determine whether rare plant species would benefit from protection from grazing by white-tailed deer after removal of Amur honeysuckle.

Measurement of false Solomon's seal also revealed a potential link between the impact of white-tailed deer on native plants and invertebrates. Lower abundance of spiders in plots accessible to deer likely was related to reduced height and leaf number of false Solomon's seal

because of reduction in availability of web attachment sites (Uetz 1991). Whether specific families of spiders, especially web builders, were more affected by deer than other families was not addressed in the present study, but such an effect could influence species diversity by selecting for non-web building species, such as wolf spiders (Lycosidae; Uetz 1975).

Both white-tailed deer and Amur honeysuckle increased compaction of forest soil, but this effect was lessened after two years of exclusion of white-tailed deer and removal of Amur honeysuckle. Despite this structural impact to the soil, neither Amur honeysuckle nor white-tailed deer altered decomposition of native leaves in forested plots. Additionally, while decomposing leaves of Amur honeysuckle supported greater microbial activity of soil in greenhouse pots than did native sugar maple (*Acer saccharum*) leaves, there was no effect of Amur honeysuckle on soil microbial activity in soil taken from invaded areas of a forest compared to uninvaded areas. While structure of of soil microbial communities and rates of litter decomposition are influenced by alterations in abundance and diversity of native plants (Hector et al. 2000), effects of white-tailed deer and Amur honeysuckle on abundance and diversity of native herbs in the present study were inconsistent. Furthermore, abundance of Collembola, which are important for initial processing of leaf litter, was similar among deer treatments, and effects of Amur honeysuckle were variable among months and years. Whether higher densities of deer or honeysuckle would alter litter decomposition is unknown. Considering the importance of litter decomposition in nutrient cycling (Seastedt and Crossley 1984), however, collection of such data could allow land managers to more easily prioritize management of white-tailed deer and Amur honeysuckle.

This study demonstrated the importance of examining effects of invasive plants and overabundant ungulates at multiple levels of organization in a variety of taxa. To understand the

breadth of effects that invasive plants and overabundant ungulates have on forest systems, future experiments could be designed to measure not only population or community-level responses, but also responses that may impact ecosystem function such as litter decomposition or nutrient cycling. Understanding ecosystem responses to invasive plants also can help reduce the incidence of unforeseen side-effects after removal of invasive species (Zavaleta et al. 2001). For example, care should be taken when removing stands of Amur honeysuckle from areas that are heavily grazed by white-tailed deer to avoid exposing plant species such as false Solomon's seal (*Mainthemum racemosa*) that may survive under Amur honeysuckle, but are highly susceptible to grazing by deer. Without this information, land managers risk attempting to restore native plant communities (e.g., species diversity) without restoring the inherent functions (e.g., litter decomposition) that maintain these systems (Zavaleta et al. 2001).

References Cited

Gorchov, D.L. and D.E. Trisel. 2003. Competitive effects of the invasive shrub, *Lonicera maackii* (Rupr.) Herder (Caprifoliaceae), on the growth and survival of native tree seedlings. Plant Ecology **166**: 13-24.

Hector, A., A.J. Beale, A. Minns, S.J. Otway, and J.H. Lawton. 2000. Consequences of the reduction of plant diversity for litter decomposition: effects through litter quality and microenvironment. Oikos 90: 357-371.

Seastedt, T.R. and D.A. Crossley, Jr. 1984. The influence of arthropods on ecosystems. BioScience 34:157-161.

Uetz, G.W. 1975. Temporal and spatial variation in species diversity of wandering spiders (Araneae) in deciduous forest litter. Environmental Entomology 4:719-724.

Uetz, G.W. 1991. Habitat structure and spider foraging. *In*: Bell, S.S., E.D. McCoy, and H.R. Mushinsky (*eds*). Habitat Structure: The Physical Arrangement of Objects in Space. Chapman and Hall. New York.

Zavaleta, E.S., R.J. Hobbs, and H.A. Mooney. 2001. Viewing invasive species removal in a whole-ecosystem context. Trends in Ecology and Evolution 16:454-459.