BIOLOGICAL CONTROL OF PURPLE LOOSESTRIFE USING *GALERUCELLA* BEETLES

by
Jessica M. Palmer

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Professor Bob Grese, Chair
Professor J. David Allan

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Abstract

Since the early 1990's, use of *Galerucella* beetles for biological control projects of purple loosestrife (*Lythrum salicaria* L.) has exhibited positive results in decreasing purple loosestrife stands and may aid in the re-establishment of native species communities.

I looked at data taken over a nine year period from permanent research plots where *Galerucella* beetles were released at the University of Michigan's Matthaei Botanical Gardens in Ann Arbor, Michigan. A path analysis was used to determine the relationship between beetle defoliation, purple loosestrife reproductive success, and native plant composition. I also compared data between years to determine where, in the course of the biocontrol program, purple loosestrife reaches its lowest density and native species begin to show signs of recovery. I found that purple loosestrife height mediates the effect of beetles on the number of plant inflorescences and that when inflorescence number and height decrease there is an increase in the floristic quality index of the native community. Sites with lower disturbance and less severe purple loosestrife invasions react positively to beetle introduction more quickly than sites with higher disturbance and purple loosestrife density. In the sites studied, approximately six years after beetle introduction, purple loosestrife reached an all time low, which suggests this as an ideal time to implement supplementary restoration techniques in order to further control the invasive population and aid in the restoration of native species.

Table of Contents

List of Tables

List of Figures

List of Photographs

I. Introduction

1.1 Purple loosestrife history

Purple loosestrife (*Lythrum salicaria* L.), a wetland plant of Eurasian origin, was first discovered in North America in 1814 (Stuckey 1980). By the late 1800s, wild populations of *Lythrum salicaria* had spread throughout the northeastern US. Purple loosestrife introduction to North America began with the transport of goods on ocean liners. In order to balance weight on trans-Atlantic crossing, ships would uptake water in Europe and, unintentionally, purple loosestrife seeds. These seeds were then discharged with the water when unloading the freight in America. European settlers immigrating to America were also vectors for purple loosestrife since it was traditionally used as herbal remedy for such ailments as digestive problems, skin lesions, and bleeding (Stuckey 1980). Purple loosestrife was also a common home landscaping plant because of its attractive dense purple flowers during the summer months. It also became apparent in the early 1900's that honey bees were attracted to these flowers and so beekeepers began cultivating purple loosestrife stands for their bee colonies. All of these cultivated uses helped to expand the wild population. Because of concerns over its spread as an invasive species, it is listed as a nuisance or noxious weed and/or banned from sale in over 34 states (http://plants.usda.gov/java/profile?symbol=LYSA2).

Once in North America, purple loosestrife was able to establish and spread quickly due to a lack of natural predators, diseases, and competition from other wetland plants. The expansion of purple loosestrife was also concurrent with widespread destruction and disturbance to wetland habitats throughout North America. It is an opportunistic colonizer, and studies have suggested that disturbed areas are prime targets for purple loosestrife invasion (Rachich 1999, Morrison 2002) and that purple loosestrife can also out-compete native wetland dominants (Weihe and Neely 1997). Purple loosestrife affects wetland ecosystems in various ways: it has the ability to out-compete native species through preferential pollination (Grabas and Laverty 1999, Brown *et al* 2002), alters phosphorus levels (Emery and Perry 1996) and nitrogen nutrient content and organic matter to its benefit (Fickbom 2006), and can affect growth of aquatic invertebrates (Gardner 2001). With each plant producing upwards of 2.7 million seeds (Willing and Becker 1993), which can be viable for up to three years (Thompson *et al* 1987), purple loosestrife is able to control species composition through its stores in the seed bank and slowly spreads to un-invaded sites from nearby established populations (Yakimowski 2005). Although purple loosestrife spreads mainly through seed dispersal, it also has the ability to reproduce vegetatively by re-sprouting at adventitious buds in the stem or crown. Also, an experiment done by Shipley and Parent (1991) suggests that the competitive ability of purple loosestrife is an effect of its faster germination and higher germination rates than native species. These characteristics, paired with being a long-lived perennial (a 4-5 year old plant is considered mature), aid purple loosestrife in easily becoming a dominant fixture in a wetland ecosystem.

Concerns over the invasiveness of purple loosestrife and its effect on the natural environment resulted in many experiments in removal techniques, including chemical treatments, plant replacement, water manipulation, prescribed fire, and mechanical methods. All, however, have shown to have little or no significant effects on purple loosestrife populations (Thompson *et al* 1987, Mal *et al* 1992, Maleki and Rawinski 1985, Passmore *et al* 2000, Blossey *et al* 2001, Weibe and Obrycki 2001). My research builds on past experiments on the control of purple loosestrife by examining a specific biological control method in a southeast Michigan wetland.

Biological control (*biocontrol*) is defined as the human manipulation of a non-native plant's predators in order to manage its population. The goal is not necessarily to completely rid the area of the invasive, but rather to regain control of the area so that an invasive monoculture can not establish and native plant communities can dominate. Biocontrol of purple loosestrife began in the late 1980's with the selection of several insect species from the plant's native European range that were known to be effective predators. Rigorous experimentation began on five insect species: the root mining weevil, *Hyblobius transversovittatus* (Goeze); two leaf eating beetles *Galerucella pumila* (Duftschmid) and *Galerucella calmariensis* (L.); two flower-feeding beetles *Nanophyes marmoratus* (Goeze), *Nanophyes brevis* (Boheman); and a gall midge, *Bayeriola salicariae* (Kieffer), (more detail found in Malecki *et al* 1993). By 1992, use of three of these insects to control purple loosestrife had passed both US and Canadian review boards. The chosen species, the leaf-eating beetles and root-mining weevil, were found to be most successful in controlling purple loosestrife, and are suitable to a broad geographic range (Malecki *et al* 1993). The research explained here concerns a biocontrol project involving purple loosestrife and the *Galerucella* beetle, and more specifically, how this beetle reduces purple loosestrife and promotes native species recovery.

1.2 Life cycle of beetle

Photo 1. Adult *Galerucella*, taken by J. Palmer.

The two commonly used biocontrol species of *Galerucella* are so similar that they are not differentiated when conducting a field study. *Galerucella* beetle adults are 3-6mm long and have two dark stripes down the sides of their light brown body (Photo 1). Their lifecycle includes a larval stage and several instars. In the months of April and May, over-wintering adults emerge from the soil and begin feeding on purple loosestrife leaves. After several days, they begin reproducing, and females lay bundles of 2-20 eggs under leaves, along leaf axils, or low on the stem. In late May or June, larvae emerge and feed on the loosestrife. Only three weeks later, they migrate down into the soil where they undergo several instars and eventually emerge as full adults. After feeding until late September, the new adults move back into the soil to over-winter and start the cycle over again the following spring.

1.3 Questions and Hypotheses: Development and Goals of Study

Matthaei Botanical Gardens (MBG), is located in Ann Arbor and Superior Townships just northeast of Ann Arbor, Michigan, on a 120 hectare parcel of land that was acquired by the University of Michigan in 1957 (http://www.lsa.umich.edu/mbg). The property contains many different ecosystems including hardwoods, conifer forest, *Carex* spp. marsh, open fields, river woods, and bogs (Wilson 1958).

Soon after incorporation of the Matthaei Botanical Gardens into the University of Michigan, Willow and Parker Ponds were dredged out of what seems to have been a wet prairie, as suggested by photos taken during at the time (B. Grese, personal communication, 2006), (Photo 2 and 3). Planting notes from 1962 show the purchase and landscaping of the Willow Pond perimeter with *Lythrum salicaria* cultivar species. As the stands expanded and knowledge of purple loosestrife's harm to ecosystems grew, MBG began looking into biocontrol as a means to manage the population. In 1995, Brian Klatt, then Associate Director of MBG, began rearing *Galerucella* beetles for release and set up a total of thirty permanent research plots in three sites near Willow Pond. My research will build on the data collected since these plots were established.

The first objective of my study was to determine the effects of beetles on the reproductive success of purple loosestrife and, in turn, the native plant community. Leaf herbivory has been shown to have the greatest impact on purple loosestrife success by reducing shoot height, reproductive success, and biomass (Hunt-Joshi *et al* 2004, Schat and Blossey 2005). Because *Galerucella* spp. is primarily a leaf-feeding beetle, any damage that it may have on purple loosestrife reproduction comes through leaf defoliation. With damage to leaves and shoots, purple loosestrife will spend more energy on healing than


Photo 2. Dredging of Willow Pond circa 1960, photo taken by Charles Cares.


Photo 3. Willow Pond after completion, photo taken by Charles Cares (early 1960's).

on producing inflorescences, thereby limiting its reproductive success. Over time, a reduction in seed release can positively affect the native plant community by increasing the number of native species and the floristic quality index (FQI). FQI is a method that uses coefficients of conservatism (C) to determine the floral rarity of a site (Herman *et al* 2001), or the probability of a plant's presence in an unaltered, pre-settlement site. This method has been found to be a useful and effective tool in Great Lakes coastal wetlands (Bourdaghs *et al* 2006). The coefficient value can range from 0 to 10 and values for Michigan species can be found in Herman *et al* (2001); values for species found in this project can be found in Appendix A. Higher values are given to species that would have been expected in pre-settlement conditions and the lowest values to species not indicative of a remnant community. FQI is determined by using the formula $FQI = \overline{C} \times \sqrt{n}$, where $\overline{C}$ is the mean coefficient of conservatism of species in the site and n is the total number of species. While FQI does not have a specific range, Michigan sites with a FQI less than 20 have little or no quality significant of remnant communities and a FQI greater than 50 exemplifies a very rare biodiversity of Michigan flora (Herman *et al* 2001).

Data from over a nine year time period will contribute to understanding the dynamics of a community under biocontrol efforts. Collecting data from the MBG test plots, I hypothesize that beetle-incurred leaf defoliation will decrease the reproductive success of purple loosestrife and improve native species diversity.

The results of my research will inform future restoration projects that consider employing similar control measures. Ecological differences in the three sites chosen for this project allow for considerations about biocontrol's effect on sites in differing stages of purple loosestrife invasion and native plant assembly. This is helpful in indicating which sites are best for biocontrol programs. And, since data from this project was collected over nine years, we can see where in that time period beetles establish and have the most effect on purple loosestrife, thus suggesting *when* supplemental restoration techniques may be useful for added control and site restoration.

II. Methods

2.1 Site Descriptions:

Three sites at MBG were chosen for this study. All are located in an area described as a *Carex* sedge meadow (Wilson 1958). Current native dominant species found in the sites include *Carex lacustris* Willd., *Tyhpa latifolia* L., *Solidago* spp., *Aster puniceus* L., and *Eupatorium maculatum* L. Although the sites are near each other (Figure 1), the ecology of each differs in species composition, disturbance levels, *Lythrum salicaria* dominance, and water levels.

Site 1 is a relatively flat area bounded by sloping hills to the south and west, a man-made hiking trail to the east, dense trees and shrubs to the north, and had a 1997 FQI of 6.30. *L. salicaria* is highly dominant here and disturbance from foot traffic is low. Standing water is rarely present although the soil may stay saturated until the late summer months. Site 2 is contained in a strip along the southern edge of Willow Pond. Purple loosestrife dominance has fluctuated over time and disturbance in high due to the proximity to the MBG Visitor's Center, picnic areas, and paths. The FQI of the site in 1997 was 4.16. Although along the pond's edge, the plots are not located in wet areas. Site 3 is also a flat area but has standing water divided by small sediment hills. Water is always present and at times flows slowly. The west edge of the site is a short steep hill leading to the MBG entrance drive and the rest of the site is surrounded by dense shrubs and trees. The purple loosestrife population has a low density and the seclusion of the site from the

Visitor Center and parking areas contributes to its low disturbance. This site also has the highest 1997 FQI, 9.37.

Photo 4. Willow Pond in 2004, photo taken by MBG staff.

Figure 1. Map of Willow Pond at the Matthaei Botanical Gardens, including study sites and layout of plots.

2.2 Monitoring protocol

Galerucella beetles were released in the spring of 1997 and monitoring began that fall. Monitoring was conducted for the next nine years (except for 2005 due to lack of staff), producing a wealth of yearly data that has allowed for longitudinal analysis. Different individuals collected the data over the length of the study and therefore the methods were standardized in order to avoid bias.

Each of the three sites contains ten randomly placed 1x1 meter plots. Monitoring was conducted biannually and conformed to the 1997 version of B. Blossey's (Cornell University) *Galerucella* and purple loosestrife monitoring protocol (PDF files may be found at www.invasivespecies.net). Spring beetle monitoring occurred in late-May as over-wintering adults emerged and began reproducing and feeding on purple loosestrife. One-minute time periods were given to count each beetle life stage—egg, larvae, or adult—and final counts were recorded. Eggs were counted as clusters of eggs, not as individual eggs. Fall plant diversity monitoring was done mid-August once the majority of beetle feeding had subsided and all plants were in full bloom. All plant species found within the plots were recorded and percent cover for each species estimated. Species list for 1997 and 2006 can be found in Appendix A. Individual purple loosestrife plants were counted and the five tallest measured. On the five tallest purple loosestrife plants, the number of inflorescences (PLI) was counted, the terminal inflorescence measured (PLT), and the number of florets (PLF) in the center 5cm of the terminal inflorescence was counted.

Note that fall monitoring in 2006 was done in mid-September while all other years it was completed in mid-August. Plot 21 and 22 in Site 3 were destroyed in 2005 when a large tree fell on top of them and therefore were not included in the 2006 monitoring sessions and are not displayed on the area map (Figure 1).

2.3 Statistical Analysis:

SPSS (version 15.0) was used for all statistical analyses. To analyze the effect of beetles on the reproductive success of purple loosestrife, I used a mediated mixed model and linear regressions. This analysis allows for consideration of the effects of time, which is an important aspect of my study. Beetles (B) was used as my X-variable and purple loosestrife inflorescence numbers (PLI), purple loosestrife floret numbers (PLF), and purple loosestrife terminal florescence length (PLT) are used as the reproductive success variables (Y). Mediator variables (M) are represented by purple loosestrife height (PLH) and purple loosestrife cover (PLC), (Figure 2). The path analysis derived from the mixed models highlights any significant relationships between beetles (X) and reproductive success variables (Y) as well as the same relationships once a mediator variable (M) is included (Figure 3). Because the path analysis can only proceed through three steps, I used linear regression to determine effects of purple loosestrife reproduction (PLI) on the native community (FQI). Regressions were plotted on Q-Q plots to ensure normality.

To determine differences in loosestrife characteristics and native species composition between sites at the onset of the project (1997) and the final year (2006), I used analysis of variance (ANOVA). Although Q-Q plots or boxplots of both raw data and transformed data did not suggest normality or equal variances, ANOVAs are robust against these violations when the sample sizes are equal, as is the case for my data (Site 1, n=10; Site 2, n=10; Site 3, n=10). Tukey's t-test was used to find which sites were significantly different from each other. I also used regression to look at yearly lag effects on several variables. I did this by comparing data of one variable to the data of a different corresponding variable the following year. Variables used and data comparisons done are shown in Table 1.

All statistical analyses used an alpha of 0.05.

Table 1. Dependent variables are shown as columns and the independent variable that was tested for its predictive ability is in the row marked with an "X".

	Beetle Year X+1	PLH YearX+1	PLI YearX+1	FQI YearX+1
Beetle YearX		X	X	
PLH YearX				X
PLI YearX				X
FQI YearX				

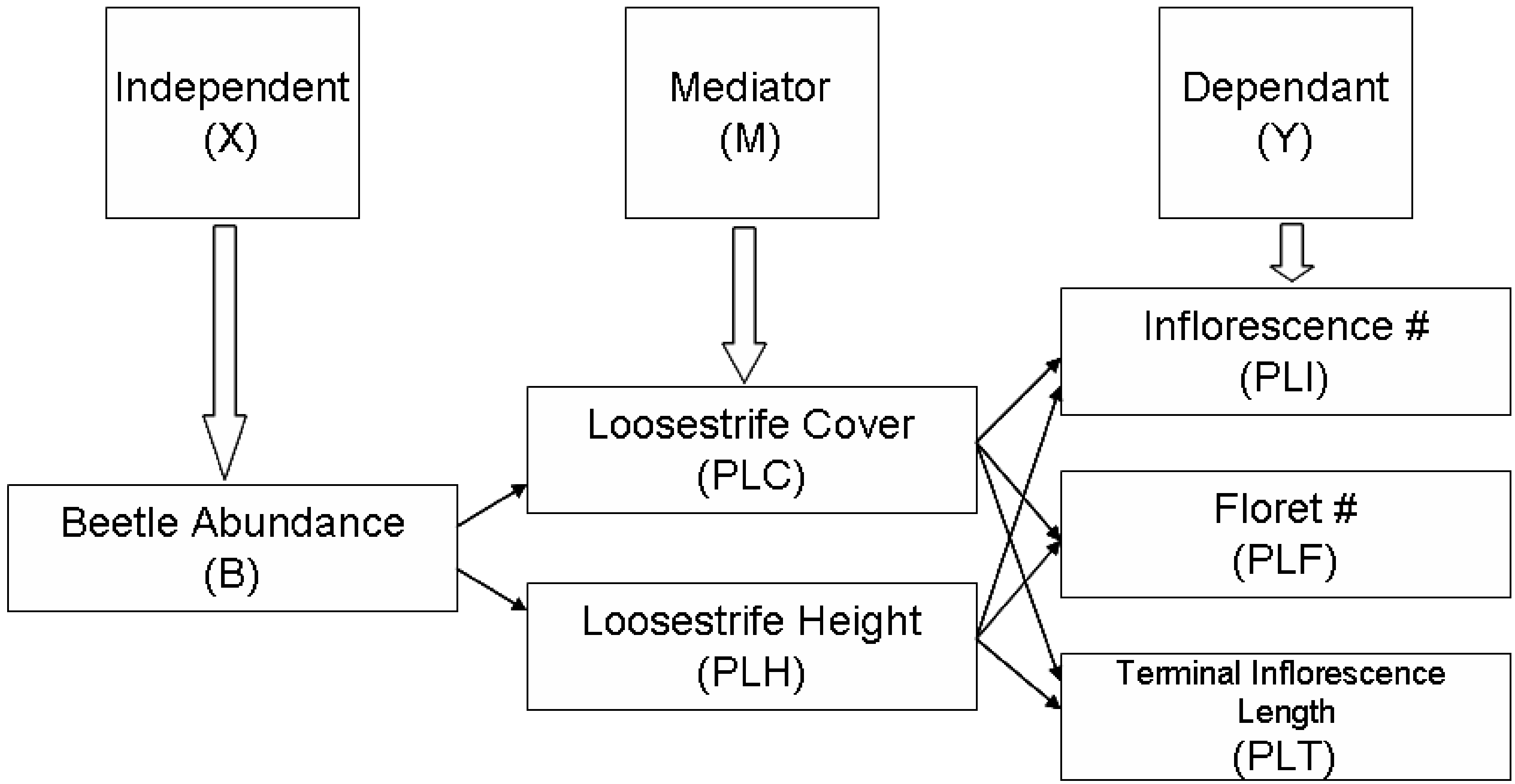

Figure 2: Path analysis model followed for mediated mixed model.

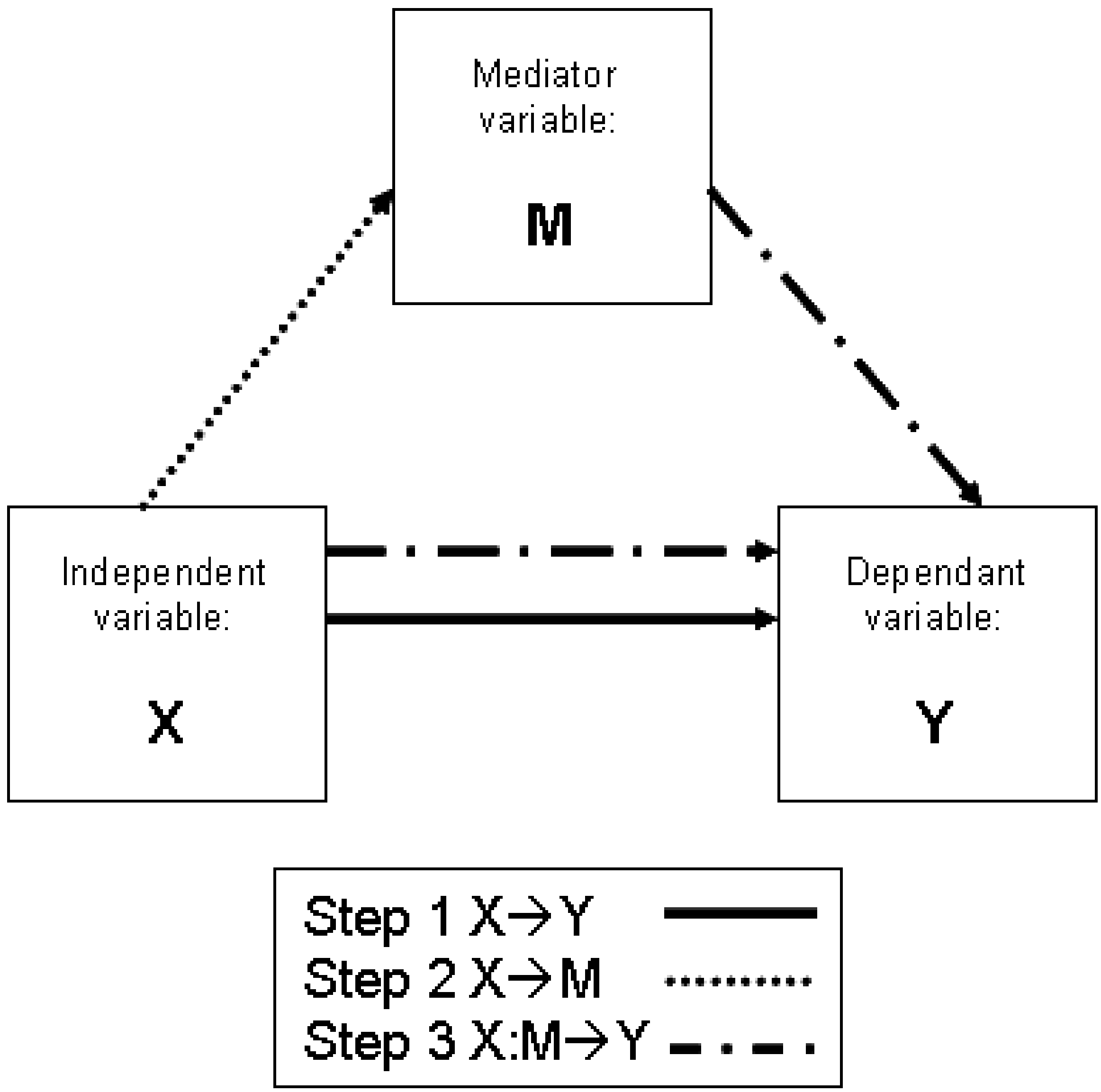

Figure 3: Model exhibiting Steps 1-3 for mediated mixed model.

III. Results

Linear graphs show an overall decrease in purple loosestrife height, cover, and inflorescence numbers (Figure 4), while there is an increase in number of native species and FQI (Figure 5) for the entire research area.

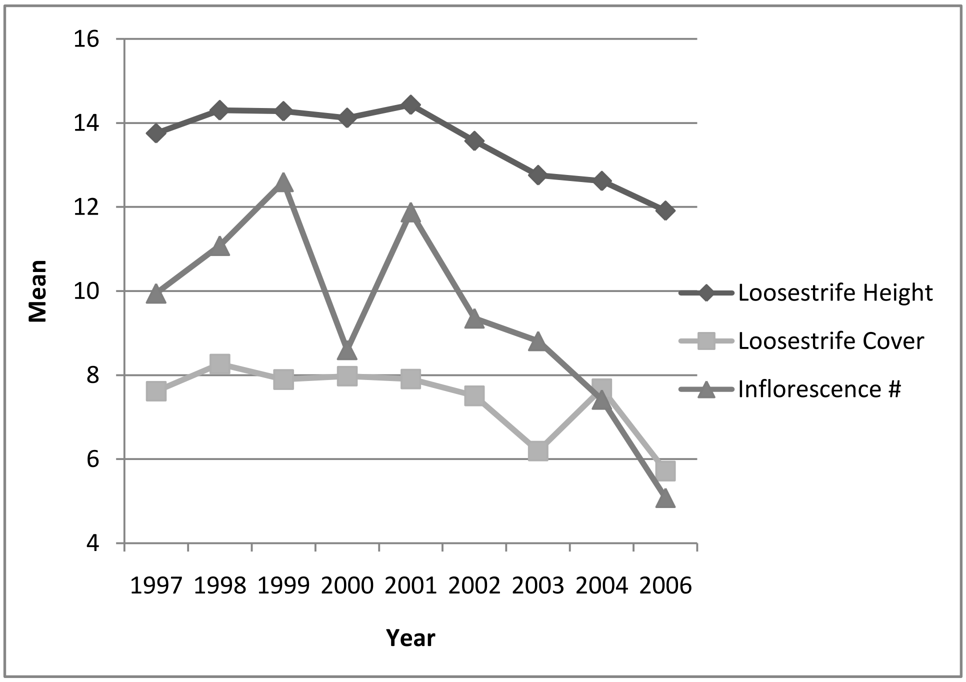

Figure 4. Trends in purple loosestrife height, cover, and inflorescence numbers over the period of the study show a decrease in all three variables. Note Y-axis intervals.

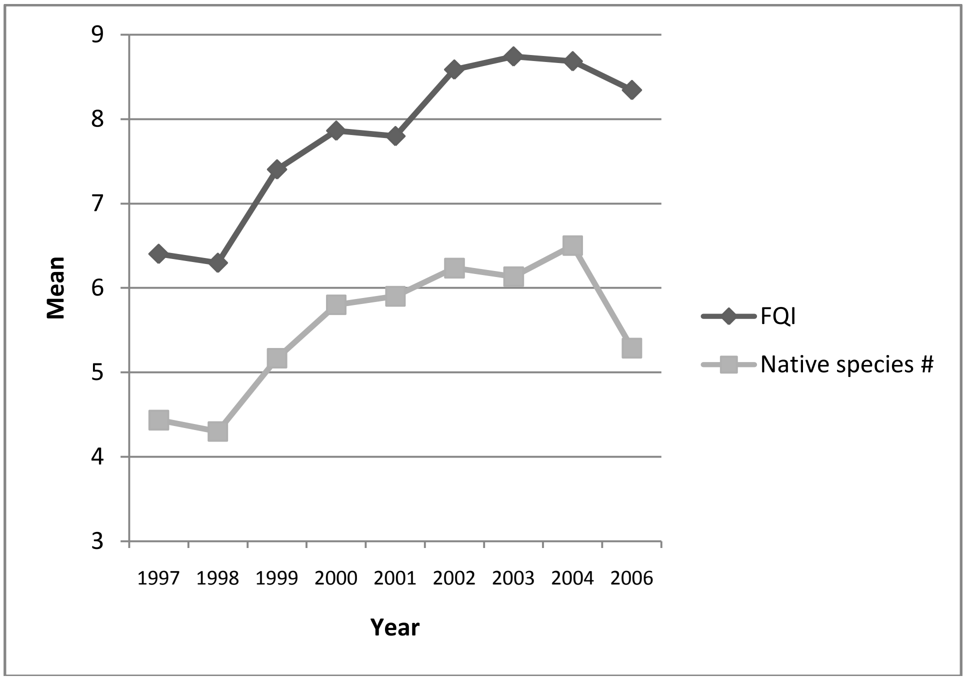

Figure 5. Trends in FQI and native species numbers show an overall increase in these two variables over the time of study. A decrease in recent years may be due to late Fall monitoring in 2006 so that all species were not recorded. Note Y-axis intervals.

3.1 First analysis: Mediation and regression

After conducting step 1 of the mediation model, purple loosestrife inflorescence numbers (PLI) was the only variable that showed a significant ($p = 0.000$) relationship to beetle abundance. Neither purple loosestrife floret numbers (PLF) nor purple loosestrife terminal inflorescence length (PLT) was significant ($p= 0.127$, $p= 0.865$, respectively). Step 2 of the mediation gave a significant relationship between beetle abundance and purple loosestrife height (PLH) ($p= 0.000$) and purple loosestrife cover (PLC) ($p= 0.026$). This required that two models were run—one for height mediation and one for cover mediation. After Step 3, the height mediation model (Figure 6) kept the significance between the beetles and PLI ($p= 0.042$) and showed significance between PLH (H) and PLI ($p= 0.000$). Therefore, the height model is a partial mediation. After completing the cover mediation model (Figure 6), the significance between beetle and PLI was lost (p= 0.774) and there was no significance between PLC (C) and PLI (p= 0.530). The cover model therefore shows no mediation.

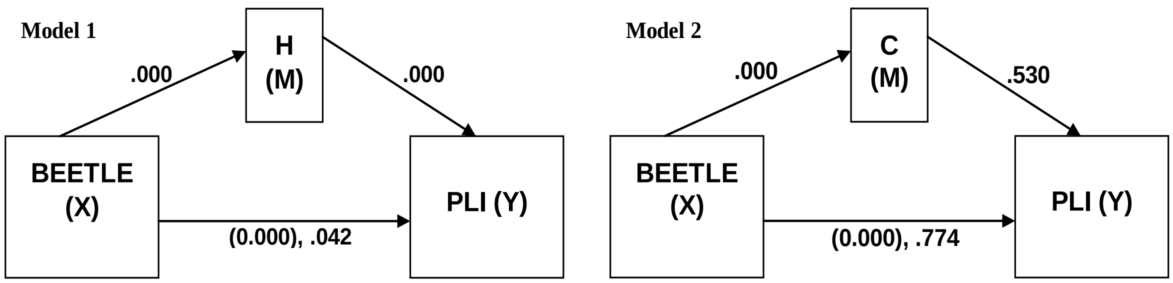

Figure 6. Linear Mixed Models: Model 1 depicting the path analysis for height as the mediator and Model 2 using cover as the mediator. Significance values are placed at corresponding relationships and the p-values in parentheses refer to p-values from Step 1.

I determined the change in FQI, PLI, and PLH over the nine year period by subtracting the 1997 value from the 2006 value. I wanted to see if when a plot had a sharp decrease in PLI or PLH over the time period, there was an increase in the FQI. The research plots were then graphically plotted, comparing FQI and PLI, and FQI and PLH. I assumed that plots shown in Quadrant II (-X, Y) suggest a decrease PLI/PLH and increase in FQI. I also used linear regression to determine any significance. Results for the linear regression do not show much significance; however, graphical representations of the change in variables from 1997 to 2006 are able to describe certain trends. FQI had no significant relationship with PLI (p= 0.935) but the comparison did suggest a trend where FQI increased with lower PLI (Figure 7). Significance was found between FQI and PLH (p= 0.011) and also had a trend where reduction in PLH allowed for an increase in FQI (Figure 8). Regressions for native and PLI and native and PLH showed no significance (p= 0.312 and p= 0.209 respectively) and neither relationships showed an obvious graphical trend.

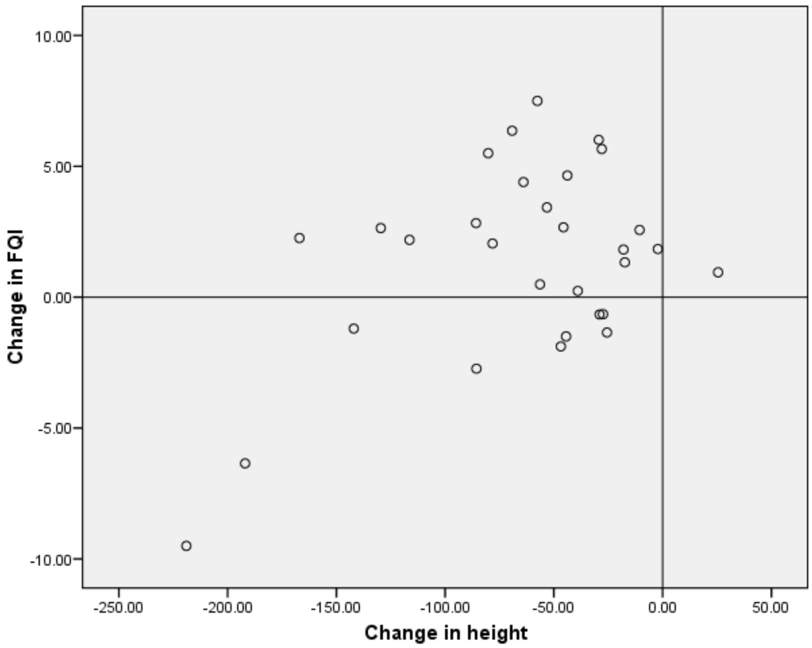

Figure 7. Each dot represents a research plot. More than half of the plots show that as the height decreases, there is evidence that the FQI in those plots increases.

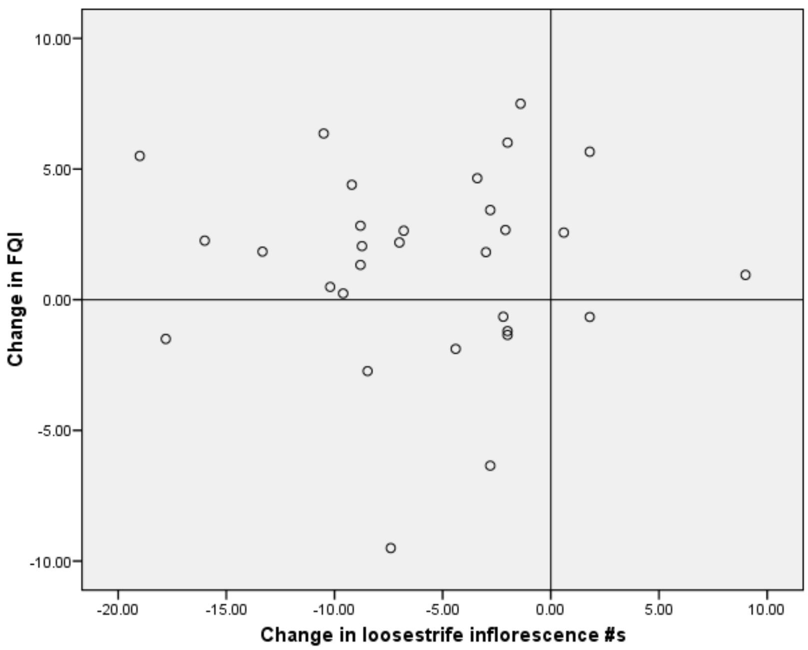

Figure 8. Again, each dot is a research plot. Half of the plots show a weak trend that as inflorescence numbers decreases, the FQI increases.

3.2 Second analysis: ANOVAs and regression

FQI: Both Site 1 and Site 2 have a significantly lower FQI than Site 3 in 1997 (p= 0.001 and p= 0.000 respectively). In 2006, Site 2 had a significantly smaller FQI than Sites 1 and 3 (p= 0.049 and p= 0.004 respectively).

Height: In 1997, there is no significant difference between the average height of purple loosestrife of the three sites (p= 0.845). By 2006, the purple loosestrife in Site 3 is significantly shorter than in Site 1 (p= 0.027).

PLI: In 1997, there is no significant difference between sites in purple loosestrife inflorescence numbers (p= 0.660). By 2006, Site 3 has significantly lower numbers of inflorescences that site 2 (p= 0.013).

PLF: The onset of the project showed no significant difference between sites in purple loosestrife floret numbers (p= 0.612). In 2006, Site 2 and 3 have significantly less floret numbers than Site 1 (p= 0.001 and p= 0.000 respectively).

PLT: In 1997, there is no significant difference between sites (p= 0.219). By 2006, Site 2 and 3 have significantly shorter terminal inflorescences than Site 1 (p= 0.000 for both).

Analysis of beetle and PLH found that the only significance was between beetle population in year 2003 and purple loosestrife height in 2004 (p= 0.000), (Figure 9). This was true as well for PLH and FQI (p= 0.000) although between years 2002 and 2003, the p-value was close to significance (p= 0.066), (Figure 10). Beetle to PLI showed close significance only between the years 2000 and 2001 (p= 0.052), (Figure 11). PLI 1999 and FQI 2000 were significant (p= 0.000), (Figure 12).

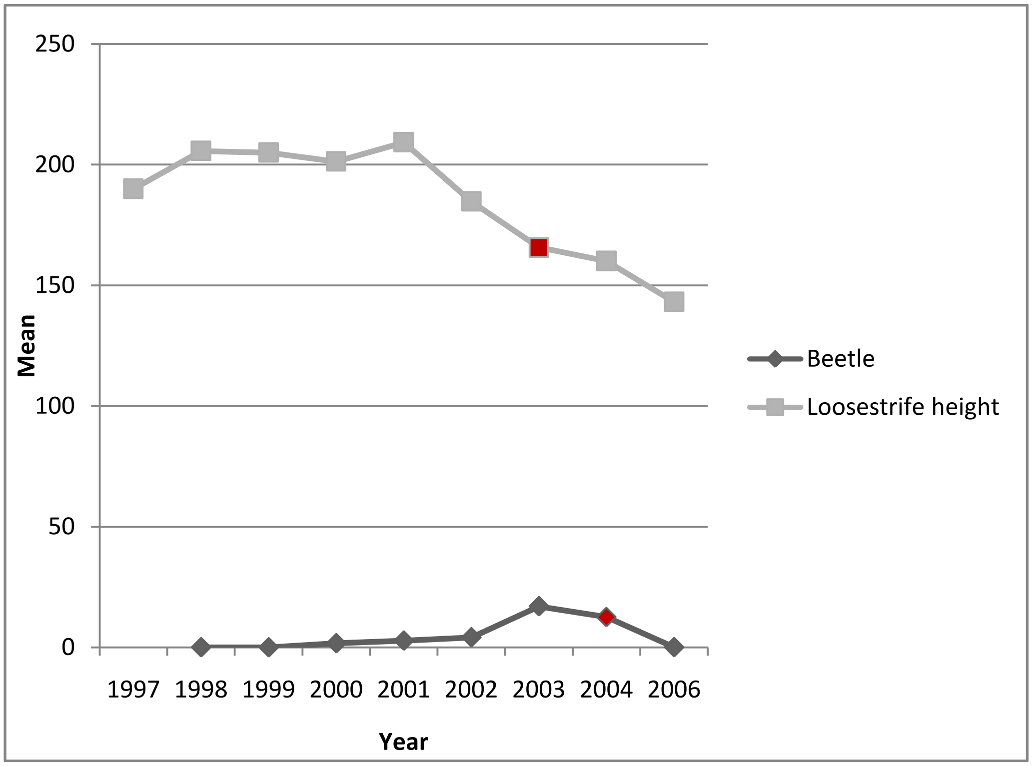

Figure 9. Beetle 2003 and PLH 2004, (p=0.000) represented in graph by red markers.

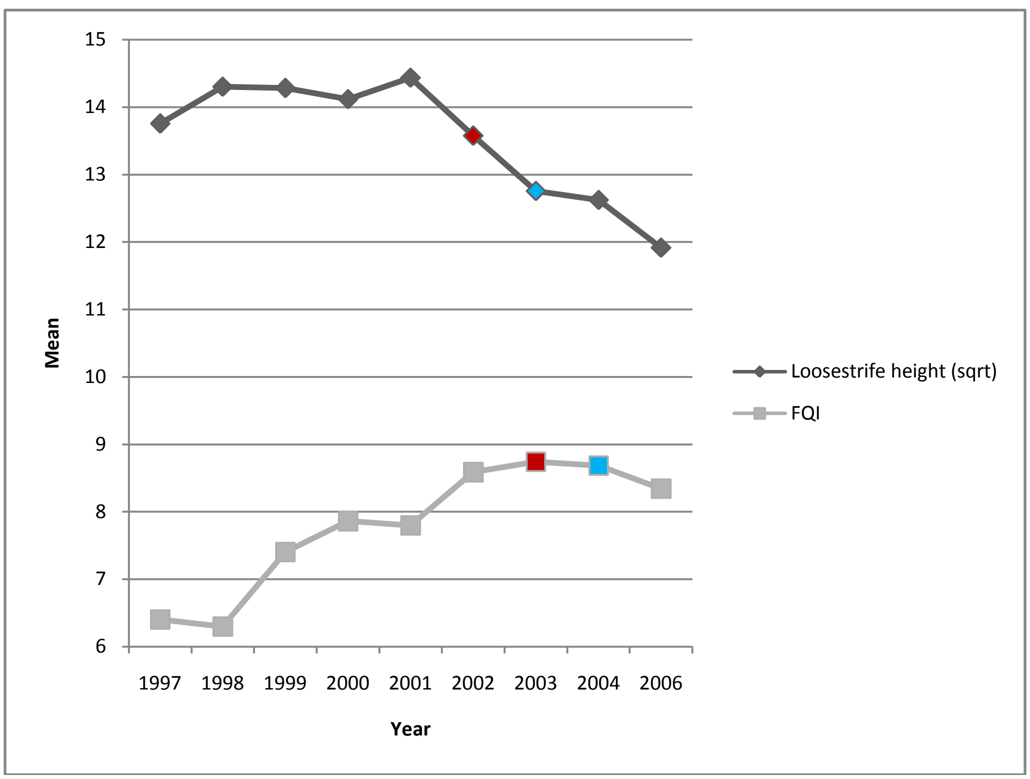

Figure 10. PLH 2002 and FQI 2003, (p=0.066) represented by red markers, PLH 2003 and FQI 2004, (p=0.000) represented by blue markers.

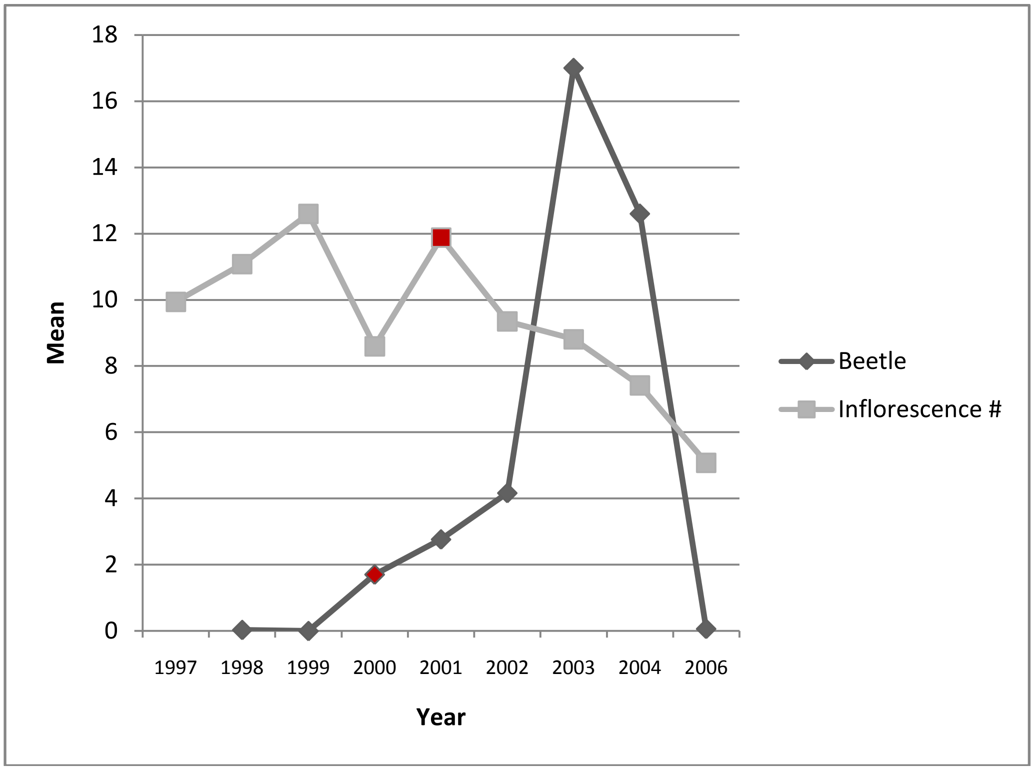

Figure 11. Beetle 2000 and PLI 2001, (p=0.052) represented by red markers.

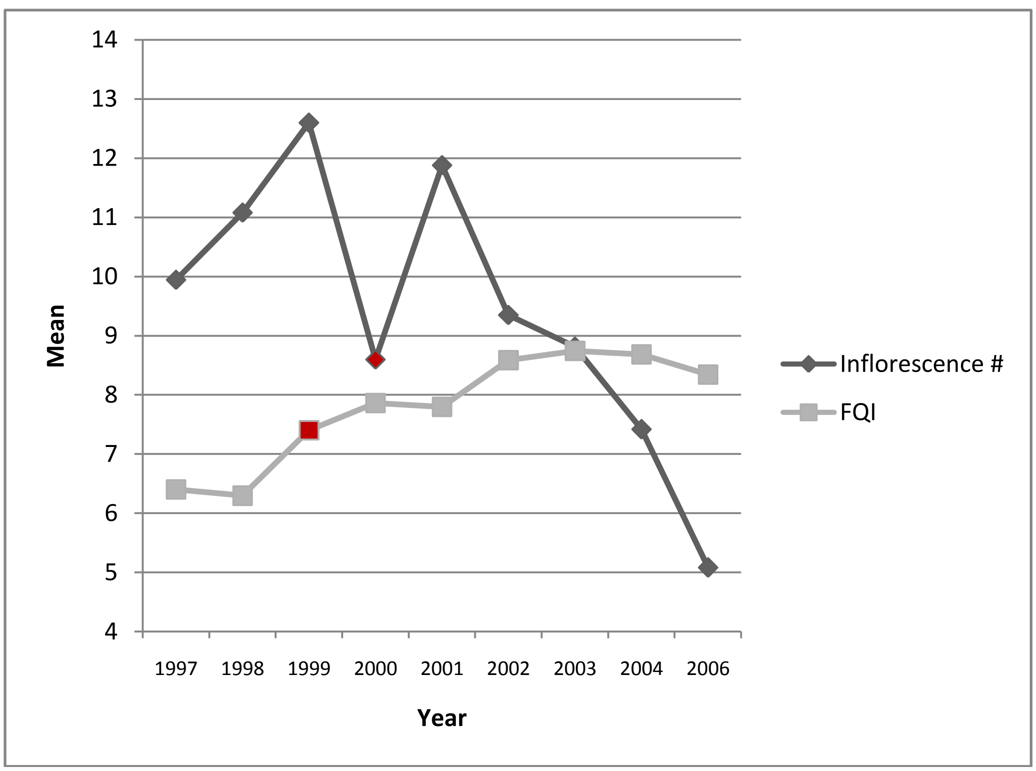

Figure 12. PLI 1999 and FQI 2000, (p=0.000) represented by red markers.

IV. DISCUSSION

Katovich *et al* (1999) suggests that *Galerucella* beetles have the potential to curb the competitive nature of purple loosestrife and aid in the restoration of a native community. This project introduced beetles to control purple loosestrife by reducing its reproductive success in order to allow the native plant community to re-establish.

When analyzing the overall study area, the mediation models hint that beetles have a significant effect on purple loosestrife reproductive success by affecting the number of inflorescences produced. *Galerucella* beetles are able to directly and indirectly (through a mediator) affect these numbers. When height is significantly reduced by beetle predation, the number of purple loosestrife inflorescences is also significantly reduced. Cover was not found to be a mediator in the path analysis model—beetles were able to directly affect PLI without first affecting purple loosestrife cover. Overall, this suggests that when faced with intense beetle damage—especially damage that reduces height—the plant must reallocate its energy from producing floral parts to repairing damage and replacing lost leaves and stems. It is suggested by other studies that insect herbivory is able to affect the inflorescence numbers (Schat and Blossey 2005), number of flowers per flower head (Hamback 2001, Denoth and Myers 2005), and decrease male flower parts and production (Lehtila and Strauss 1999) through damage done to leaves and shoots. Fitness and health of mature plants can be negatively affected by damage to cotyledons (Hanley and Fegan 2007) and leaf herbivory (Vallius and Salenon 2005). Female reproduction is decreased when leaves and buds are damaged as attractiveness to pollinators drops (Mothershead and Marquis 2000). Lower seedset and fitness, caused by insect herbivory, of a community dominant can boost other community species. In my study, relationships between PLH and PLI to FQI were not significant but the quadrant graphs show that as purple loosestrife height and inflorescence numbers decrease, the floristic quality of the study area increased. However, when analyzed with native species numbers, PLI and PLH were not significant nor had any immediate trends. This may have been because the Fall 2006 monitoring was delayed until later in the season when some species may have already died and were neglected in the count. The fact that FQI increased but native species did not could be because of the late monitoring of 2006 or because species with a higher coefficient of conservatism (used in determining the FQI) have replaced species with a lower coefficient—or even a combination of both possibilities.

Although none of the plant characteristics studied was significantly different in 1997, by 2006, Site 3 had lower height, less inflorescence and floret numbers, and shorter terminal inflorescences. This site also had the highest FQI at the start and at the end. From this data, areas with ecology similar to Site 3 have the greatest biocontrol success. Because this site had high native diversity, which may lead to higher levels of plant species competition, purple loosestrife may not have been able to establish. This kind of relationship is known as the diversity-invasibility hypothesis and it tests whether or not areas with high native diversity are less susceptible to invasion. While there has been controversy on the diversity-invasibility hypothesis, Knops *et al* (1999) were able to show that areas with high species diversity were less susceptible to invasion. Their results also supported the relationship that high diversity means high resource use, and therefore lower invasibility. In addition, in areas where multiple plant groups have been depleted, indicating a decrease in diversity, the invasion success of aggressive species rises (Rinella *et al* 2007). Site 1 and 2 seem to be in a transition stage where neither is exhibiting significance towards purple loosestrife control and native establishment since the variables used to determine this change are not in agreement within each site. However, these data show that areas with high native diversity and lower levels of disturbance respond fastest to biocontrol efforts. Areas with higher disturbance, lower diversity in the native community, or denser purple loosestrife populations still show a positive response to beetle introduction but the response may take longer to occur. In addition, the reduction in purple loosestrife may actually facilitate the increase in other aggressive species. For example, observations in Site 2 show a shift from purple loosestrife dominance to narrow leaved cattail (*Typha angustifolia*) dominance over the study period and could have caused a setback in native species recovery. This suggests that as beetles begin to affect the purple loosestrife population, care should be taken to watch for encroachment by other aggressive species such as cattail. Even though cattail is native, Houlahan and Findlay (2004) concluded that exotic species were no more likely to dominate areas than native species and the key to a diverse community is to discourage spreading of any dominant species.

Comparing data across years, I did not find a linear path from beetles to height/inflorescence numbers to FQI as demonstrated by the path analysis in the mediation model. I found that beetles affected purple loosestrife plants (PLI and PLH) a year later than when the PLH and PLI affected FQI. This is opposite of what I expected and signifies that more research is needed to understand better the year to year effects between independent and dependent variables so that predator-prey relationships for this association may be better understood.

V. Conclusion

Combining biocontrol techniques with other restoration methods can be more efficient in curbing purple loosestrife colonies than a single technique (Katovich *et al* 1999). Biocontrol projects are predicted to have more success when using more than one insect predator (Malecki *et al* 1993). Wetland restoration managers using biocontrol could consider using other methods such as seeding or planting plugs around the five year mark of using *Galerucella* beetles if the site does not already have a diversity of other native species poised to fill in the void left by purple loosestrife as it declines. My project shows that around that time, purple loosestrife populations hit a low point and may be further depleted by introducing supplemental restoration techniques that encourage competition from other plant species. Also, in late summer, when adult beetles are in diapause, purple loosestrife has a period of relief from predation so it can recover and have a late season growth spurt and allocate energy into the root mass for the winter (Katovich *et al* 1999, Grevstad 2006, Yakimowski *et al* 2005). Supplemental methods during this time of year could halt recovery and energy storage, to the detriment of the purple loosestrife population.

My project just touches on the complexities inherent to biocontrol projects, and while the data presented here has been collected over nine years, it is limited in its ability to completely describe patterns and determine the mechanisms that create them. We do see that *Galerucella* beetles are able to reduce purple loosestrife populations; while the beetle's ability to boost native species diversity is feasible, the path toward that goal is not clear. Height has a greater effect on the native community than expected under the original hypothesis. Reducing the height of purple loosestrife plants may be the first step in native comeback by allowing in light through gaps in the shorter vegetation. Once native species establish and purple loosestrife reproductive success in limited, growth of those native species may be able to surge. Data from my project supports that beetle biocontrol negatively affects purple loosestrife and that at a certain time after introduction purple loosestrife reaches a low point before showing signs of resurgence. Therefore, introduction of *Galerucella* beetles, in conjunction with other timely restoration methods, can help re-establish native communities and quell monotypic stands of purple loosestrife.

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Appendix A
Species List

Scientific	Common	CC	1997	2006
Acer negundo	BOX ELDER	0		x
*AGROSTIS STOLONIFERA**	CREEPING BENT		**X**	
Amphicarpaea bracteata	HOG-PEANUT	5	x	x
Apios americana	GROUNDNUT; INDIAN-POTATO	3	x	
Apocynum cannabinum	INDIAN HEMP; HEMP DOGBANE	3	x	x
Asclepias syriaca	COMMON MILKWEED	1	x	x
Aster puniceus	SWAMP ASTER	5	x	x
*BERBERIS SPP**	JAPANESE BARBERRY			**X**
Bidens connatus	PURPLE STEMMED-TICKSEED	5		x
*BRASSICA RAPA**	FIELD MUSTARD; TURNIP			**X**
Calamagrostis canadensis	BLUE-JOINT GRASS	3		x
Campanula aparinoides	MARSH BELLFLOWER	7		x
Carex lacustris	SEDGE	6	x	x
Carex stricta	SEDGE	4	x	x
Chelone glabra	TURTLEHEAD	7		x
Chelone obliqua	RED TURTLEHEAD	9		x
*CIRSIUM ARVENSE**	CANADIAN-THISTLE			**X**
*CONIUM MACULATA**	POISON HEMLOCK			**X**
Cornus amomum	SILKY or PALE DOGWOOD	2	x	x
Cornus foemina	GRAY DOGWOOD	1	x	x
Cornus stolonifera	RED-OSIER DOGWOOD	2	x	x
Epilobium coloratum	CINNAMON WILLOW-HERB	3		x
Equisetum fluviatile	WATER HORSETAIL	7		x
Eupatorium maculatum	JOE-PYE WEED	4	x	x
Euthamia graminifolia	GRASS-LEAVED GOLDENROD	3	x	x
Galium aparine	ANNUAL BEDSTRAW	0		x
Galium asprellum	ROUGH BEDSTRAW	5	x	x
Geum canadense	WHITE AVENS	1		x
Glyceria grandis	REED MANNA GRASS	6	x	x
Glyceria striata	FOWL MANNA GRASS	4		x
Impatiens capensis	SPOTTED TOUCH-ME-NOT	2	x	x
Iris versicolor	WILD BLUE FLAG	5		x
Juncus dudleyi	DUDLEY'S RUSH	1	x	x
Juncus tenuous	PATH RUSH	1		x
Leersia oryzoides	CUT GRASS	3	x	x
Lemna minor	SMALL DUCKWEED	5	x	x
Lobelia syphilitica	GREAT BLUE LOBELIA	4		x
Lycopus americanus	COMMON WATER HOREHOUND	2	x	x
Lysimachia ciliata	FRINGED LOOSESTRIFE	8		x
*LYTHRUM SALICARIA**	PURPLE LOOSESTRIFE		**X**	**X**
Mentha arvensis	WILD MINT	3		x
*MENTHA SPICATA**	SPEARMINT		**X**	
*NASTURTIUM OFFICINALE**	WATERCRESS		**X**	
Parthenocissus quinquefolia	VIRGINIA CREEPER	5	x	
Phalaris arundinacea	REED CANARY GRASS	0	x	x
*POA PRATENSIS**	KENTUCKY BLUEGRASS		**X**	**X**

Polygonum hydropiper	WATER-PEPPER	1		x
Pycnanthemum virginianum	COMMON MOUNTAIN MINT	5	x	
Ranunculus hispidus	SWAMP BUTTERCUP	5		x
*RHAMNUS FRANGULA**	GLOSSY BUCKTHORN			**X**
Ribes americanum	WILD BLACK CURRANT	6	x	
*ROSA MULTIFLORA**	MULTIFLORA ROSE		**X**	**X**
Sagittaria latifolia	COMMON ARROWHEAD	1	x	x
Sambucus canadensis	ELDERBERRY; COMMON ELDER	3	x	x
Senecio aureus	GOLDEN RAGWORT	5	x	x
Solidago altissima	TALL GOLDENROD	1	x	x
Stellaria longifolia	LONG-LEAVED CHICKWEED	5	x	x
Symplocarpus foetidus	SKUNK-CABBAGE	6	x	x
Thalictrum dasycarpum	PURPLE MEADOW-RUE	3	x	x
Toxicodendron radicans	POISON-IVY	2	x	
*TYPHA ANGUSTIFOLIA**	NARROW-LEAVED CAT-TAIL			**X**
Typha latifolia	BROAD-LEAVED CAT-TAIL	1	x	x
Uniola latifolia	WILD-OATS	10		x
Urtica dioica	NETTLE	1	x	x
Verbena urticifolia	WHITE VERVAIN	5	x	
Viola cucullata	MARSH VIOLET	5	x	x
Vitis riparia	RIVERBANK GRAPE	3	x	
	Total native		36	48
	Total invasive		6	9

* listed as invasive
CC= coeff. of conservatism