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Influence of Stem Cutting and Glyphosate Treatment of *Lonicera maackii*, an Exotic and Invasive Species, on Stem Regrowth and Native Species Richness

A.L. McDonnell[1], A.M. Mounteer[1,2,3], H.R. Owen[1,4], and B.L. Todd[1]
[1]Eastern Illinois University, Charleston, IL 61920
[2]Brigham Young University—Idaho, Rexburg, ID 83440
[3]current address: Purdue University, West Lafayette, IN 47906
[4]author for correspondence (hrowen@eiu.edu)

ABSTRACT

Lonicera maackii (Rupr.) Herder (Caprifoliaceae), Amur honeysuckle, is an exotic and invasive species in the United States that has quickly overtaken disturbed habitats in the eastern and midwestern United States, as well as in Ontario, Canada. A reduction of light due to its dense canopy, extended growing season compared to native species, and production of numerous basal sprouts allow *L. maackii* to outcompete its native counterparts. Eradication of this species can be difficult and time-consuming. This research was undertaken to identify how *L. maackii* influences species diversity and species re-establishment and to determine an efficient and effective eradication method. A study was designed to determine if *L. maackii* inhibited species diversity, if the removal of *L. maackii* would increase species diversity by reopening the canopy, and if mechanical removal or mechanical removal coupled with glyphosate treatment could be used effectively for its long-term eradication. It was found that *L. maackii* removal increased species diversity, and mechanical removal coupled with the application of glyphosate is an effective and relatively simple method for eradicating *L. maackii*, while mechanical stem removal alone simply delayed its growth.

INTRODUCTION

Lonicera maackii (Rupr.) Herder (Caprifoliaceae), Amur honeysuckle, was introduced into the United States in 1897 from Asia because of its ornamental properties (attractive flowers, fruit, and fragrance) and ability to provide habitat for wildlife [Minnesota Department of Natural Resources (MDNR), 2003; Hutchinson and Vankat, 1997; Luken et al., 1997; Luken and Thieret, 1996]. *L. maackii* is an erect, deciduous shrub that can range anywhere from 2 to 6 m in height when mature. Its invasion into forests and disturbed areas began as it escaped cultivation. *L. maackii* has quickly overtaken disturbed habitats, especially in the last few decades, and has also been known to invade open areas with little vegetation that are not disturbed (Collier and Vankat, 2002; Luken and Thieret, 1996; Trisel and Gorchov, 1994). It is now found in at least 24 states throughout the eastern and mid-western U.S. and in Ontario, Canada (Hutchinson and Vankat, 1997; Trisel and Gorchov, 1994).

L. maackii seedlings are likely to establish themselves in forest understories and in open areas with little vegetation (MDNR, 2003; Hutchinson and Vankat, 1997; Luken and Theiret, 1996). Seeds were shown to germinate readily in both light and dark environments (Luken and Goessling, 1995). High germination rates and aggressive plant growth also contribute to the competitiveness of *L. maackii* with native species. In addition, maturing plants were shown to adapt and grow well in areas with different light intensities (Luken, 1988). Growth and reproduction are aggressive and rapid. Initial fruits are produced when the plant becomes woody, within three to five years after seedling establishment (Luken and Thieret, 1996).

The invasiveness of Amur honeysuckle also is a function of its extended growing season, ranging from early March to mid November (Trisel and Gorchov, 1994). Early expansion of *L. maackii* leaves has negative effects on native plants (especially spring ephemerals) giving it a distinct competitive advantage (Hoffman and Kearns, 1997; Trisel and Gorchov, 1994). Leaves of *L. maackii* were found to be photosynthetically active for a significantly longer time than three native species (*Acer saccharum* Marsh., *Asimina triloba* (L.) Dunal. (Pawpaw), and *Lindera benzoin* (L.) Blume.) (Trisel and Gorchov, 1994). Also, leaves are retained on *L. maackii* plants longer (until mid November) than most native plants, due to lack of herbivory and biological control agents (MDNR, 2003). Biological controls are not known for *L. maackii*, and leaf damage recorded at the end of the growing season was shown to be lower for *L. maackii* than for other native, woody species (Batcher and Stiles, 2003; Trisel and Gorchov, 1994). The lack of biological control agents, as shown on a related species, *Lonicera japonica* Thunb. var. japonica, illustrates how important biological agents are in maintaining plant communities (Schierenbeck et al., 1994). Although it has been suggested that bush honeysuckles (including *L. tartarica* L., *L. maackii*, and *L. morrowii* Gray) may produce allelopathic chemicals, it has not been documented (MDNR, 2003).

Both early leaf expansion and extended growing season are two factors that affect species richness by creating a dense canopy, which is negatively associated with native and non-native species richness (Collier and Vankat, 2002; Gould and Gorchov, 2000; Hoffman and Kearns, 1997; Hutchinson and Vankat, 1997). This dense canopy may inhibit native species by depleting soil moisture and nutrients and reducing light availability (Collier and Vankat, 2002; Gould and Gorchov, 2000; Hoffman and Kearns, 1997; Hutchinson and Vankat, 1997). Species richness and abundance and tree seedling abundance were shown to be significantly greater in areas without an Amur honeysuckle canopy cover compared to areas where *L. maackii* had produced a dense canopy (Collier and Vankat, 2002; Hutchinson and Vankat, 1994). Results of a study done showing the effects of canopy cover of *L. maackii* on native annuals illustrated that annual plants, particularly shade intolerant species, declined in the presence of a canopy cover (Gould and Gorchov, 2000). One study demonstrated increased light availability (about 10% of full sun) for plants where *L. maackii* shrubs were removed, compared to approximately 1% of full sun light availability where shrubs remained (Luken et al., 1997). Light availability significantly and positively increased overall plant density; however, increasing light availability for plants may not only encourage native species to grow, but also promote non-native, invasive species, such as *Allaria petiolata* (Bieb.) Cavara & Grande (garlic mustard) (Luken et al., 1997).

Large quantities of red berries are produced by *L. maackii* (Luken and Thieret, 1996; Ingold and Craycraft, 1983). The main agent of seed dispersal is birds that consume the red berries (Ingold and Craycraft, 1983).

L. maackii abundance has the ability to negatively alter forest structures and native herbaceous populations (Hutchinson and Vankat, 1997). It has also been shown that the longer *L. maackii* shrubs persist in forests, species richness and abundance of tree seedlings decreased (Collier and Vankat, 2002).

The flowers of *L. maackii* are initially white and become yellowish with age and emit an attractive fragrance (MDNR, 2003). Many homeowners desire this plant in their gardens because it is one of the most aesthetically pleasing species among honeysuckles (Bean, 1973). While the shrub is still planted as a decorative landscaping specimen, there are efforts to eradicate it. Eradication can be a difficult and time-consuming process because of its invasive and anti-herbivory qualities. The two most widely documented methods for successful eradication have been burning and glyphosate (RoundUp™) application to either whole shrubs or cut stems (Batcher and Stiles, 2003; MDNR, 2003). However, there is a potential hazard with using burning as an eradication method, especially in residential areas. Another eradication method that has been used with some success is mechanical removal of entire shrubs; however, this method is particularly disruptive to residential landscapes. Therefore, in residential areas, applying glyphosate to a cut stem is a more appropriate method. Any of these methods would take more than a year to completely prevent its re-establishment because the seed bank in the surrounding soil may be quite extensive due to *L. maackii*'s abundant seed production (Batcher and Stiles, 2003). In this study, a practical approach to the removal of *L. maackii* was taken to determine whether the presence of *L. maackii* inhibits diversity, whether the opened canopy would enable the once abundant wildflowers to re-establish themselves, and the efficacy of applying concentrated glyphosate directly to cut stems compared to mechanical stem removal only.

MATERIALS AND METHODS

This study was conducted on a residential area in Charleston (Coles Co.), Illinois (39°28'N, 80°10'W) from mid May to mid July in 2003 and 2004. Three 20 m transects (approximately 5m between transects) were positioned running east and west (Fig. 1). Data from all honeysuckle shrubs within 2m of all transects was recorded. In May 2003, *L. maackii* stems in transects A and B were mechanically removed (either by saw or pruning shears, where appropriate) 10 cm above the soil surface. In addition, all newly cut stems within 2 m of transect A were treated with an 18% solution of glyphosate [n-(phosphonomethyl)glycine] by painting the solution directly onto cut stems. Transect C remained untreated (i.e. no removal, no glyphosate application).

An initial inventory of mature honeysuckle, including number and height of mature shrubs, number of stems per shrub, and stem diameter was determined prior to any removal. An inventory of species diversity and abundance was recorded in alternating 0.5

m^2 quadrats (40 quadrats/transect). After the mechanical removal of honeysuckle stems (May 28, 2003), six weekly inventories (June 6 to July 9, 2003) of each transect were conducted to monitor any re-growth of honeysuckle stems and growth of understory plants. Species inventory was also recorded weekly in the second year (May 26 to July 8, 2004). Stem numbers and lengths of mature *L. maackii* shrubs in transect A (mechanical removal coupled with glyphosate) and transect B (mechanical removal alone) were recorded weekly in both 2003 (May 28 through July 9) and 2004 (May 26 through July 8). Species richness was tested using both univariate analyses of variances (ANOVAs) and Tukey tests with SPSS 13.0 (SPSS, Inc., Chicago, IL). Weekly measurements also were taken of light intensity (µmol/m^2/sec; 13 cm above soil surface) and soil temperature (°C; 5-8 cm below soil surface) at three locations along each transect. Significant differences in light intensities and soil temperatures were also analyzed using ANOVAs with SPSS 13.0. All analyses were conducted with an alpha level of 0.05.

RESULTS

Stem data

The initial inventory of mature, woody honeysuckle shrubs in all three transects was taken in the first week of both years (Table 1). No new shoots sprouted in 2004 on honeysuckle shrubs that had bee treated with glyphosate in 2003. Stem number and stem length also were recorded to determine the extent of re-growth after mechanical removal alone. In the first year, shoots started to emerge after three weeks on cut honeysuckle stems in transect B (no glyphosate). Most of these shoots were buds, so no stem lengths were recorded until week four. The second year of data revealed that stem number did not increase greatly, but stem length increased almost threefold (Fig. 2). Biomass (as determined by average stem number multiplied by average stem length) increased by more than three times of the amount of total re-growth recorded at the end of the first year (432) as compared to indicated biomass in the second year (1469).

Species richness

Species quantity and type were recorded over a span of seven weeks for both years and the total average frequency was calculated to show richness of different species per transect (Table 2). Some species were difficult to identify as seedlings at the beginning of data collection for both years, until the plants grew and became more distinguishable. These small seedlings were recorded in the "miscellaneous seedlings" category until they were able to be identified in subsequent weeks.

In the first year, statistical analyses were conducted to determine density of three species of plants [*Viola pratincola* Greene (violets), *Acer negundo* L. var. negundo (boxelder), *Lonicera maackii*] and species richness. Each species was tested using a univariate analysis of variance. *Lonicera maackii* and *Acer negundo* seedlings and species richness were also analyzed from the second year of data. *Viola pratincola* showed a significant difference in all weeks (all weeks showed a probability value=0.000, df=2, 117, all r^2 values <0.340) for all of the transects, including its initial inventory. Because the difference in abundance of *V. pratincola* between transects was present from the beginning of the study, it was deemed a non-useful indicator of species re-establishment and therefore they were not analyzed in the second year. In contrast, since boxelder and Amur honeysuckle seedlings were uniformly distributed amongst all transects in the beginning of the study, they were deemed useful indicators of species re-establishment.

In the first year, there were not any significant differences of *Acer negundo* seedlings between transects for all weeks under study (Fig. 3). Week three of the second year was the first week that illustrated any significant differences between transects. Both cut transects, A and B, showed significantly greater abundances of boxelder seedlings compared to transect C (p=0.003, df=2, 117 and p=0.036, df=2, 117, respectively). However, in this week there was not a significant difference between transect A and transect B (p=0.682, df=2, 117). Also, week four of the second year showed seedling abundance to be significantly greater in transect A compared to transect B (p=0.016, df=2, 117) and transect A compared to transect C (p=0.000, df=2, 117). However, there was not a significant difference between transect B and the control (p=0.503, df=2, 117). Seedling abundance was greatest in transect A (glyphosate) compared to the other transects for all weeks in the second year as shown in Figure 3.

The number of *Lonicera maackii* seedlings were not significantly different in the three treatments during any of the test weeks in the first year (p>0.05 for all weeks, df=2, 117) (Fig. 4). In the second year, however, *L. maackii* seedlings in transect A were significantly greater than seedlings in transect C for all weeks except week two (p<0.046 for all weeks that were significantly different, df=2, 117) (Fig. 4).

Lonicera maackii seedlings in week four of the second year were analyzed. The number of seedlings in transect A compared to transect B were significantly greater (p=0.000, df=2, 117). The number of seedlings in transect A compared to the control were also significantly greater (p=0.000, df=2, 117). The number of seedlings in transect B compared to the control were not significantly different (p=0.709, df=2, 117).

Before the honeysuckle stems were cut, there were no significant differences in species richness between treatments. On June 25, 2003 (week 5) there was a significant difference between transect A and the control in this week (p=0.007, df=2, 117) (Fig. 5). However, there were not significant differences between transect A and transect B (p=0.051, df=2, 117) and transect B and the control (p=0.754, df=2, 117). Significant differences observed in the first year did not persist throughout all the weeks of this study. Week five of the second year showed there to be significant differences between all transects (p=0.000, df=2, 117) (Fig. 5). All transects showed significant differences between each other (p=0.000, df=2, 117, for all combinations). As the canopy returned (in 2004 in transect B), species richness was lower than transect A (Fig. 5).

Environmental data

Soil temperatures for both years were not significantly different between transects when analyzed in SPSS 13.0 in 2003 (p=0.089, df=2, 54), and in 2004 (p=0.231, df=2, 60). Light intensities for both years were shown to be significantly different between transects in 2003 (p=0.007, df=2, 54) and in 2004 (p=0.000, df=2, 60). Mean values for light intensities and soil temperatures are shown in Table 3.

DISCUSSION

Stem data

The initial inventory of the mature honeysuckle indicates similar plant densities and sizes of the shrubs among the three transects, thus enabling useful comparisons. Results show that *L. maackii* is a dominant species at this location. Cut stems without glyphosate application initiated new shoots within three weeks, illustrating the inefficiency of mechanical stem removal as a singular eradication method. By contrast, no stems were initiated in either the first or second years on plants cut and treated with glyphosate, suggesting that the herbicide killed those individuals.

The significant re-growth of stems in transect B (compared to the total lack of regrowth in transect A) indicates that mechanical stem removal without herbicide treatment does not eradicate *L. maackii*. In fact, overall stem lengths were much greater in the second year for transect B, illustrating that not only is mechanical stem removal without herbicide treatment an unsuccessful eradication method, it actually promotes new shoot growth that would quickly re-establish biomass (Fig. 2). Mean heights of *L. maackii* in the control transect were estimated data, thus explaining a decrease in mean heights of mature honeysuckle shrubs in this transect from 2003 to 2004 (Table 1).

Species richness

The number of *Acer negundo* seedlings was much larger in the first year than the second year. However, in the second year, although there were fewer boxelder seedlings overall, there were significant differences between all three transects (Fig. 3). Evidently, there were more boxelder seedlings in the seedbank in 2003 than 2004. Although boxelder seedlings were not noted to be significantly greater in transect A compared to transects B and C until week 4, Figure 3 shows that transect A (glyphosate) contained the greatest number of boxelder seedlings in all weeks. By week three, there were significantly fewer boxelder seedlings in the control transect than in the two removal transects, suggesting that it is an opportunistic species. This information supports the initial hypothesis that seedling abundance is greater in areas where light availability is increased due to canopy removal. Also, since seedling abundance was shown to be greater in transect A (glyphosate) than transect B for most weeks in the second year of study, it is apparent that mechanical stem removal alone (in transect B) does not increase its species richness in that area.

The number of *L. maackii* seedlings was not significantly different between all three transects in the first year. Amur honeysuckle seedlings were approximately uniformly distributed throughout all transects for the first year of data collection. A dramatic increase in seedling abundance (in both transects A and B) in the second year is illustrated by Figure 4. This increase in seedling abundance further emphasizes that increased light availability by canopy removal (or reduced canopy) positively influences seedling density. In the second year, honeysuckle seedlings were significantly greater in transect A as compared to transects B and C (Fig. 4). Seedlings likely sprouted from the seedbank and were able to survive due to the newly opened canopy that previously inhibited seedling establishment in all transects. Due to unsuccessful eradication techniques, the canopy in transect B had begun re-establishing itself (by the summer of 2004), and there were fewer seedlings in this transect, which may be due to reduced light caused by the returning canopy. The significantly greater number of seedlings in transect A indicates that more seedlings are able to grow in the absence of a canopy; however, there were some differences in the understory species between the transects where *L. maackii* was removed and the control in the first year. Data from the second year indicates that there is a significant difference in species richness between all three transects. Figure 5 illustrates that species richness was greater where there was a more open canopy. Transect A showed the greatest species richness, where transect B showed significantly less species richness than transect A (Fig. 5). Species richness in both transects A and B was significantly greater than the species richness in transect C. Our results agree with Collier's and Vankat's (2002) findings that species richness is lower where *L. maackii* develops a dense canopy. This result makes sense given the invasive qualities of *L. maackii.* It is interesting that, in the initial inventory, there were more *L. maackii* seedlings in transect C than in either A or B. However, after *L. maackii* was removed from A and B and the canopy was reopened, honeysuckle seedlings in transects A and B increased slightly (more so in the second year), while at the same time decreased in transect C over the length of the study. Figure 5 suggests that ridding a transect of its honeysuckle canopy leads to a greater species richness, and that mechanical stem removal alone (transect B) does not prohibit regrowth of the canopy, which ultimately leads to a lower species diversity. Also, since species richness was found to be greater in transect A than transects B and C (week five of the second year), and species richness was not found to be significantly different between transect B and the control in this same week, this data further shows evidence of a lower species diversity and richness in areas where Amur honeysuckle was only mechanically removed and its canopy regrowth inhibited an increase of species diversity.

Native species such as *Acer saccharinum* (L.) (silver maple), *Aster lateriflorus* (L.) Brit. (side-flowering aster), and *Impatiens capensis* Meerb. (spotted touch-me-not) were observed in the second year of data but not the first (Table 2). Table 2 also illustrates that *Aster lateriflorus* seedling density was observed to be greater in transect A (glyphosate) than both other transects. The violets increased as well, once the canopy was opened. The mean number of violets increased greatly in both cut transects in 2004. *Geum canadense* Jacq. increased in abundance in transect A in the second year compared to its mean value in the first year. This species was also more abundant in transect A compared to the other transects in the second year.

All non-native species observed the second year were observed in the first year as well (Table 2). However, *Taraxacum officinale* Weber (dandelion) was observed the second year but not in the first. Since dandelions were only noted in transect A (glyphosate), this indicates that non-native plant species are likely to invade in areas where *L. maackii* has been removed. Also, Amur honeysuckle seedlings increased dramatically in transect A in the second year, indicating yet again that non-native species are likely to invade when Amur honeysuckle's canopy is removed.

After analyzing the second year of data, it was found that species richness increased when the canopy was opened and mechanical removal alone was not a successful technique for eradication. Even after cutting and treatment with glyphosate, *L. maackii* likely persists in the seedbank. Removing sprouting seedlings by hand after removing the canopy may be a technique to remove all honeysuckle plants. Although this removal may need to be done over more than one year, it is probably one of the most successful ways to eradicate *L. maackii* completely in an area.

Environmental data

Results of this study are in accordance with previous studies that illustrated that light intensity was noticeably lower in the control compared to transects A and B in the first and second years (Hutchinson and Vankat, 1997; Luken et al., 1997). This difference indicates that *L. maackii* produces a dense canopy, shading and inhibiting herbaceous plant growth. Light intensity was higher in transect A in the second year, as compared to the other transects. Since the canopy remained opened in the second year, this could be a reason why the light intensity was greater in that transect. Also, since a large amount of regrowth appeared in transect B during the second year, its light intensity would be expected to be lower than transect A.

Soil temperatures were not significantly different between transects in the second year, however. Soil temperatures may have shown no significant differences due to low, herbaceous plants shading the ground in transects A and B.

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Figure 1. Layout of study site. The shaded area rectangle represents the area where the Amur honeysuckle shrubs were cut within two meters of transects A and B.

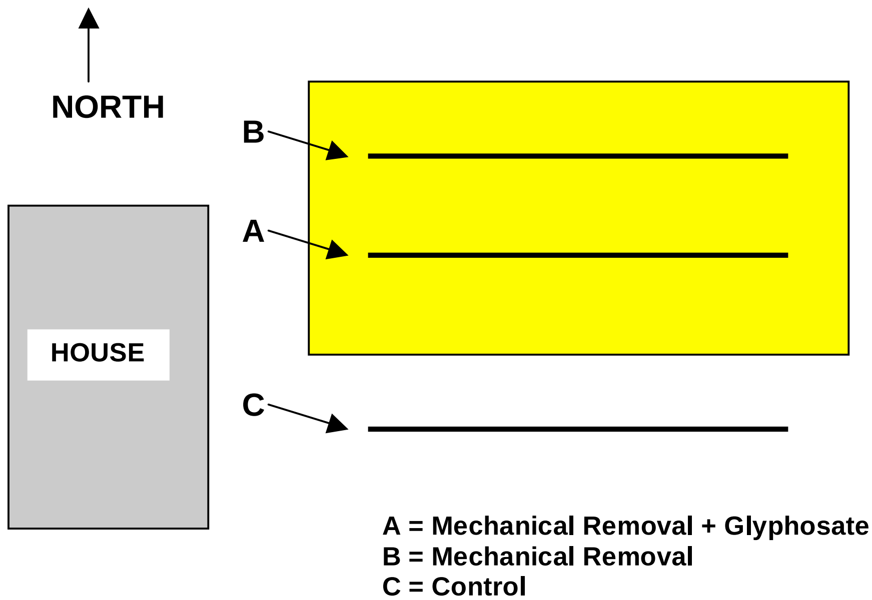


Table 1. Initial inventory of mature *L. maackii* within all three transects from the first and second years of data with standard errors.

	# of shrubs	shrubs/m^2	mean height (m)	mean # stems/shrub	mean stem diameter (mm)
2003					
Glyphosate	34.0	3.4	2.6 ± 0.16	4.4 ± 0.42	22.9 ± 1.40
Cut	23.0	2.3	3.0 ± 0.19	5.0 ± 0.51	38.1 ± 14.4
Control	30.0	3.0	3.0 ± 0.18	3.5 ± 0.37	24.0 ± 1.46
2004					
Glyphosate	0.0	0.0	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Cut	20.0	2.0	1.2 ± 0.09	14.4 ± 1.43	4.6 ± 0.26
Control	30.0	3.0	2.7 ± 0.13	3.8 ± 0.049	26.6 ± 2.76

Table 2. Overall mean abundance (plants/m^2) of understory plant species in 2003 and 2004 by transect for all weeks (Mohlenbrock, 2002).

	Glyphosate	Cut	Control
2003			
Native			
Acer negundo	6.2	6.2	4.7
*Brachythecium oxycladon**	1.9	0.4	0.4
Carex jamesii	1.6	1.1	0.1
Geum canadense	1.8	0.2	1.3
Sambucus canadensis	0.0	0.4	0.0
Smilax hispida	0.1	0.0	0.2
Toxicodendron radicans	0.7	0.0	0.5
Viola pratincola	10.9	31.7	0.7
Non-native			
*Ajuga reptans**	0.0	6.4	0.0
Lonicera japonica	0.0	0.1	3.0
Lonicera maackii	1.3	1.3	1.9
Morus alba	0.0	0.0	0.1
Rosa multiflora	0.4	0.1	0.4
*Vinca minor**	0.0	0.0	7.3
Miscellaneous seedlings	16.1	17.2	13.5
Other	0.7	0.5	0.3
2004			
Native			
Acer negundo	28.5	13.0	2.8
Acer saccharinum	5.0	1.8	1.6
Aster lateriflorus	11.0	4.5	0.0
*Brachythecium oxycladon**	5.0	0.5	0.5
Cercis canadensis	0.0	1.2	4.3
Fraxinus americana	0.0	0.5	0.5
Fraxinus lanceolata	62.5	54.5	2.5
Geum canadense	38.3	3.8	9.0
Impatiens capensis	16.3	1.0	0.0
Oxalis stricta	2.0	2.3	0.0
Phytolacca americana	0.3	0.0	0.0
Toxicodendron radicans	14.5	0.0	11.8
Viola pratincola	568.0	825.8	4.5
Non-native			
*Ajuga reptans**	0.0	2.7	0.0
Lonicera japonica	3.0	2.5	3.5
Lonicera maackii	135.3	79.8	49.0
Morus alba	2.5	0.5	2.5
Rosa multiflora	9.8	1.3	4.3
Taraxacum officinale	2.1	0.0	0.0
*Vinca minor**	0.0	0.0	14.8
Miscellaneous seedlings	66.0	28.8	49.5

*Indicates a percentage cover.

Figure 2. Average stem numbers and stem lengths in transect B (mechanical stem removal only) of Amur honeysuckle recorded in the seventh week of data collection in 2003 and 2004.

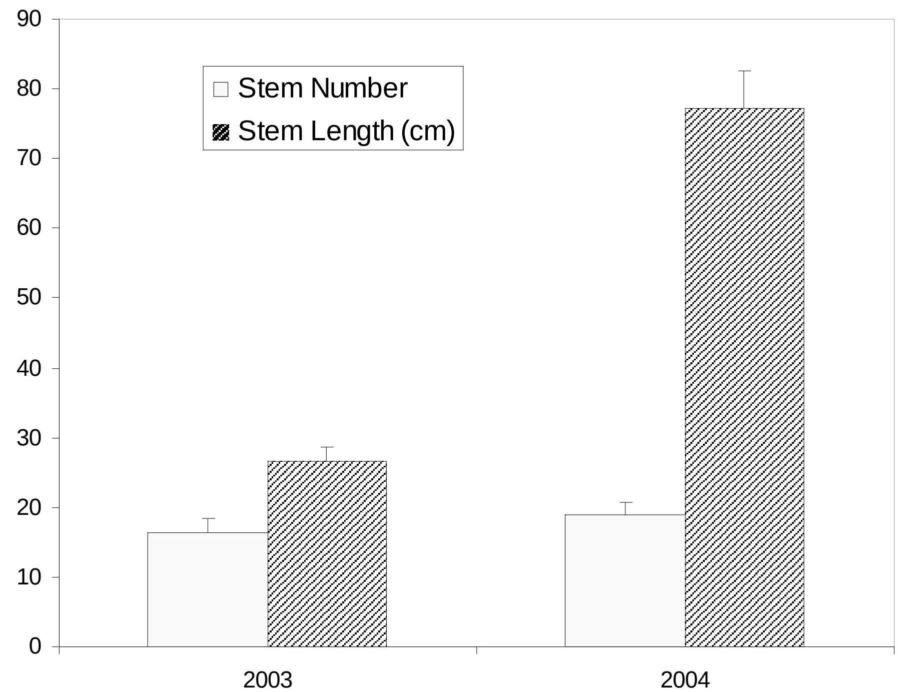


Figure 3. Mean number of *Acer negundo* seedlings per square meter in all transects.

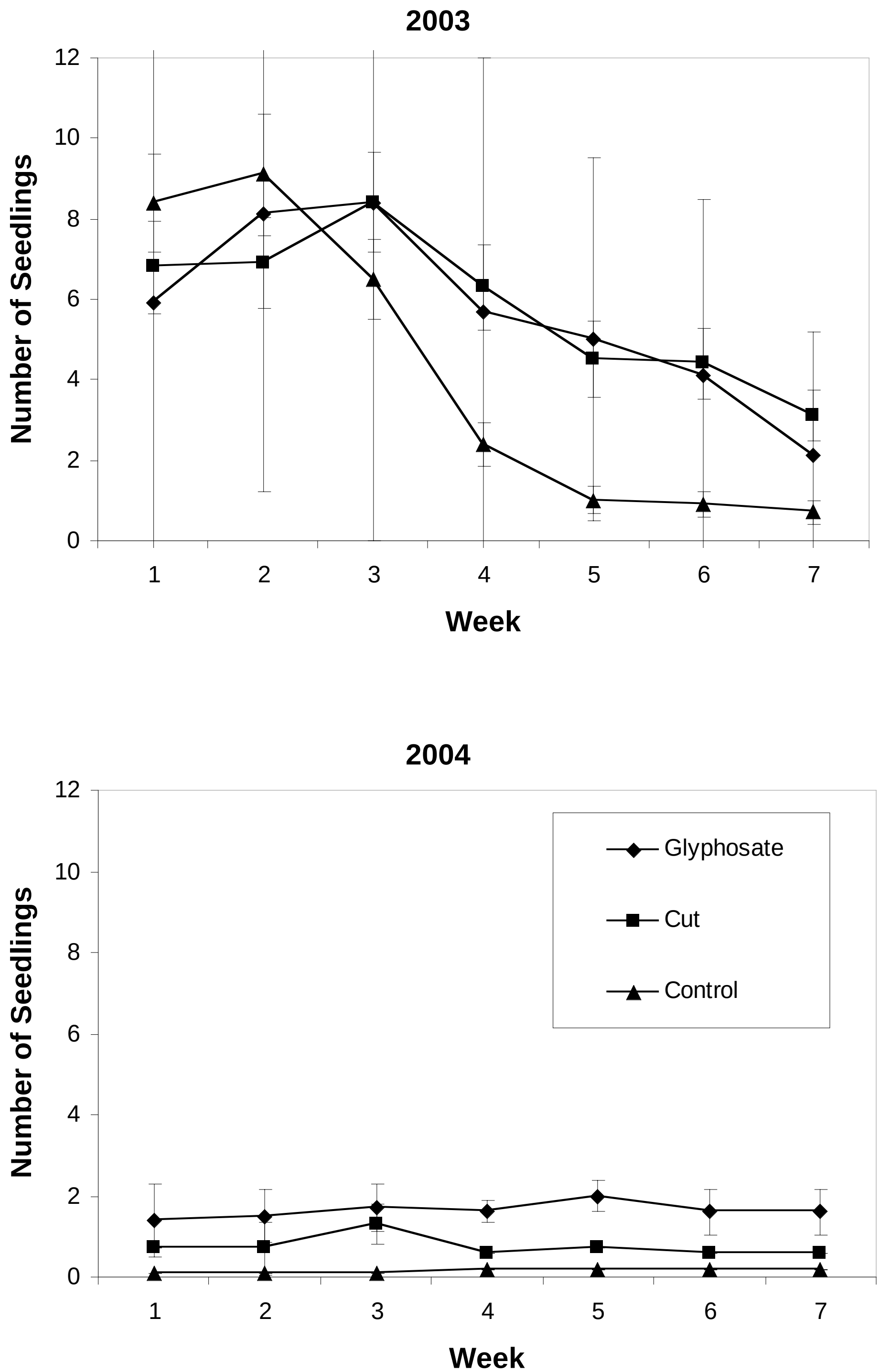
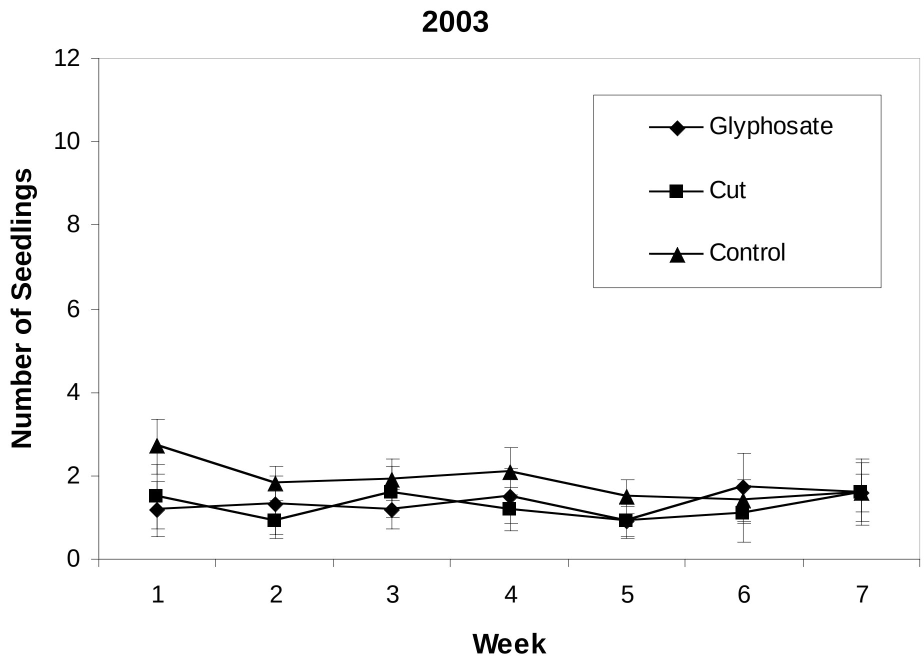


Figure 4. Mean number of *Lonicera maackii* seedlings per square meter in all transects.



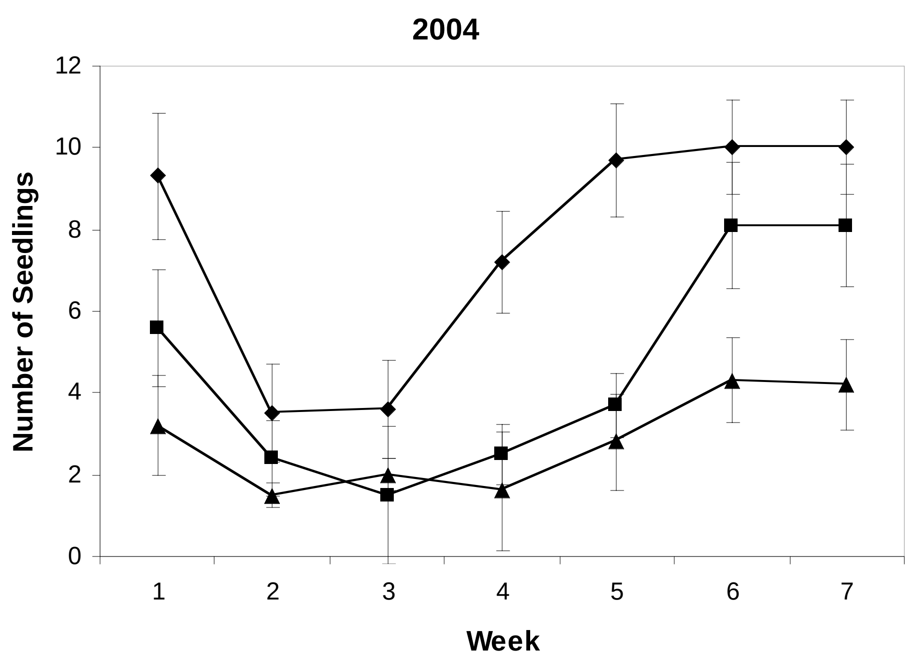
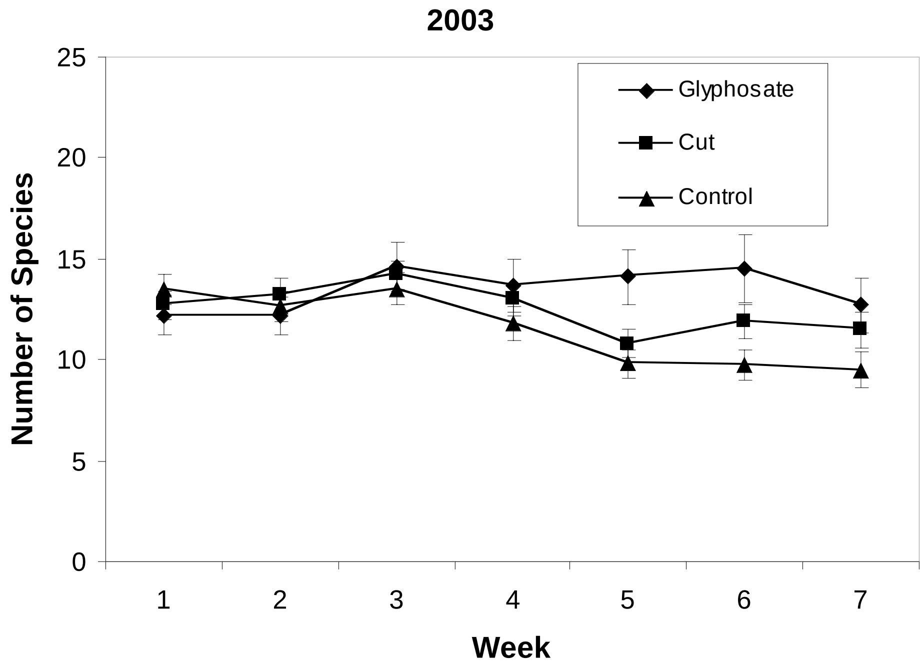


Figure 5. Mean species richness per square meter for all transects.



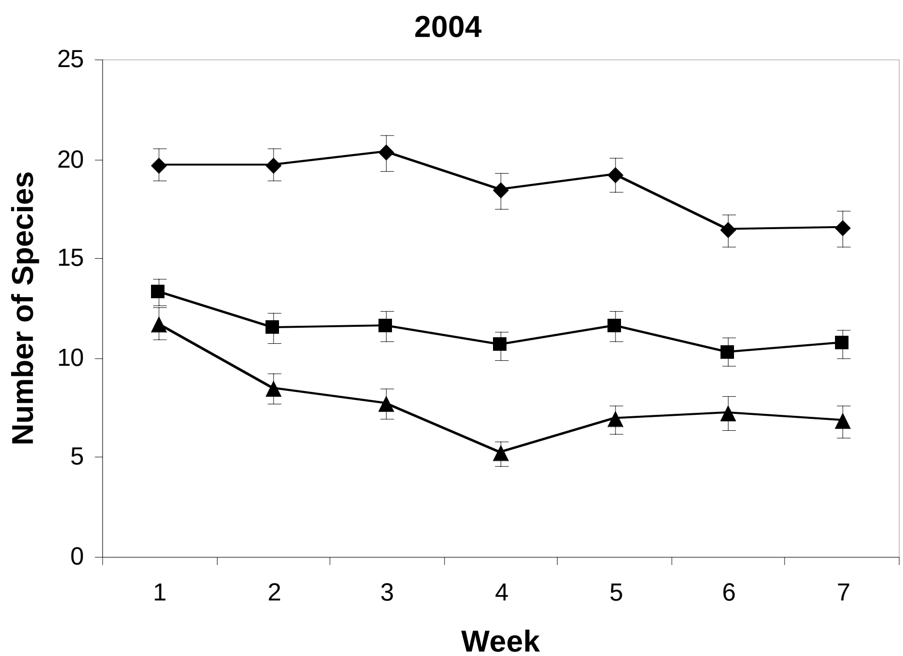


Table 3. Mean light intensities ($\mu mol/m^2s^{-1}$) and soil temperatures by week and year.

		LIGHT INTENSITY (PPFD)			SOIL TEMPERATURE (^0C)		
	week	glyphosate	cut	control	glyphosate	cut	control
2003	1	*	*	*	17.2	*	*
	2	29.0	39.0	9.0	17.4	17.4	15.7
	3	56.0	49.3	14.3	22.4	22.2	19.9
	4	63.0	53.3	15.3	23.4	24.6	21.0
	5	92.7	133.0	45.0	25.9	25.0	22.3
	6	27.0	32.0	7.7	25.9	24.2	22.3
	7	23.0	30.7	10.0	26.7	26.3	24.7
2004	1	19.7	14.0	9.7	19.4	18.9	18.3
	2	9.0	18.7	1.3	17.8	17.6	17.1
	3	57.7	21.3	13.7	21.9	21.4	20.9
	4	24.0	13.7	10.3	23.4	23.4	22.5
	5	22.3	8.0	8.7	21.5	20.9	20.0
	6	32.3	20.0	5.7	21.8	21.3	20.9
	7	24.7	13.7	8.0	22.1	21.8	21.4

* Data not available.