

# Physiologically Based Control of Invasive Asiatic Shrub Honeysuckle

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## ABSTRACT

Asiatic shrub honeysuckles (*Lonicera maackii*, *L. x bella* complex) are widespread invaders of Midwestern forests, damaging both plant and animal habitat. Techniques for the elimination of honeysuckle are not well evaluated in the literature. Stem cutting with application of 20% glyphosate to the stubs is widely used, but is not completely successful. It is also fairly painstaking, uses significant quantities of concentrated herbicide, and is purportedly ineffective in spring. Our objective was to develop control techniques which capitalized on the physiological characteristics of understory shrubs, relying on a timed cutting regime and limited or no herbicide application. In one experiment we evaluated whether cutting after leaf expansion followed by foliar spraying or cutting regrowth were effective means of control. In a second experiment we evaluated the effect of cutting height and regrowth spraying on control. Cutting near ground level just after leaf expansion followed by spraying or cutting regrown foliage with dilute (0.9%) glyphosate proved as effective as published results for stem cutting and concentrated glyphosate application. There are significant benefits to adopting early season cutting and midsummer retreatment as a control technique, particularly for private landowners.

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## INTRODUCTION

Asiatic shrub honeysuckles (*Lonicera maackii* and the *L. x bella* hybrid complex = *L. tatarica* x *L. morrowii*) are widespread invaders of forest habitats throughout eastern North America. They often achieve superabundance in forest edges and disturbed stands, excluding native plant species (Woods 1993, Gould and Gorchov 2000, Collier *et al.* 2002) and degrading wildlife habitat (Ingold and Craycraft 1983, White and Stiles 1992, Schmidt and Whelan 1999). Land managers in the Midwest are particularly likely to encounter honeysuckle invasions because the fragmented landscape offers ideal habitat for avian fruit dispersers, and provides plenty of forest edge and disturbed forest (Luken and Goessling 1995).

Honeysuckle extermination is difficult for a number of reasons. The shrub is hard to kill completely (Batcher and Stiles 2000), and resprouts readily (Luken and Mattimiro 1991). Many invaded stands are physically challenging, featuring numerous individuals (e.g., 1800 ha<sup>-1</sup>, Schulz, unpublished data) of large size (2-5 m in height). Typically, desirable

species are also present, limiting widespread herbicide application or mechanical disturbance. After populations have been exterminated, reestablishment from bird dispersed seed is a continuing concern (Luken and Goessling 1995, White and Stiles 1992).

Based on conversations with resource managers and other professionals, an array of honeysuckle control techniques have been attempted, but few are described, much less evaluated outside the “grey” literature. A few techniques are effective, although each has its drawbacks: In an appropriate situation fire can be used to control honeysuckle, although repeated fires or other secondary treatment are nearly always necessary for satisfactory results (Nyboer 1992). Foliar spraying with 1.5% glyphosate is effective when applied to mature shrubs just after flowering (Nyboer 1992), however there is an obvious practical problem treating large shrubs while minimizing overspray on nontarget species. Honeysuckles can be pulled from soft moist soils, although soil disturbance produces seed beds for weedy species and opportunities for reinfestation (IPAW 2007).

Kline (1981) originated what appears to be the most widely recommended extermination technique: cutting stems at ground level in summer (or later) and painting the stumps with concentrated glyphosate. She achieved an 89.0% mortality rate using a 1:5 glyphosate:water solution on *L. x bella* in southern Wisconsin. Reportedly this technique kills honeysuckle even in the winter dormant season (Missouri Conservation Commission 2007), although its efficacy in a broad range of situations is not documented in the wider literature. The Missouri Conservation Commission (2007) comments that glyphosate stump treatments on *L. maackii* are less effective in spring because resources are directed toward new buds as opposed to roots. Reduced effectiveness in spring was not observed by Love *et al.* (2006) working on *L. morrowii* in Pennsylvania.

Although it is part of popular practice, the choice of glyphosate as a *stump treatment* and its use throughout the year has little theoretical support. Kline’s rationale for choosing glyphosate pivoted on its benign characteristics during an era when there were few alternatives (Schulz, personal communication). Glyphosate formulated as Roundup®, Kleenup®, and other mixtures has low toxicity to mammals (*Rattus* oral LD<sub>50</sub> > 5000 mg kg<sup>-1</sup>), and is apparently benign to a wide array of *terrestrial* organisms (Monsanto 2006) (cf. Relyea 2005a,b). It is recommended for foliar application (Roundup® label directions), being readily absorbed by leaves and translocated through both xylem and phloem to meristems. Direct injection into large honeysuckle stems is effective, but this requires specialized apparatus (Hartman and McCarthy 2004). Severed vascular tissue is obviously not ideal for uptake, evidenced by the fact that concentrations for stump treatment are 10 to 20 times stronger than foliar sprays.

Glyphosate functions by inhibiting amino acid synthesis and is most effective in rapidly growing plants (Shaner 2006). Herbicide treatments in summer, fall, and later do not coincide with rapid plant growth. Growth rates of most temperate woody species are greatest in spring (Kozlowski and Pallardy 1997). In the lower Midwest, moisture stress during midsummer caused by higher temperatures and reductions in rainfall (NCDC 2007) potentially reduce physiological activity (Abrams and Mostoller 1995, Kozlowski and Pallardy 1997). Kline (1981) noted vegetative growth resumed during fall in *L. x bella* populations near Madison, WI. Based on our observations in southwestern Illinois,

this phenomenon does not generalize to populations of *L. maackii*, and may not apply elsewhere.

From the standpoint of environmental protection, expense, and practicality, additional issues deserve notice. Glyphosate is strongly adsorbed in soil, but persists a longer period (60 d) than many herbicides used in forest management (McNabb 1996). Limiting discharge of glyphosate is desirable because it is not only toxic to all plants but there is evidence that the Roundup® formulation is toxic to amphibian larvae (Relyea 2005a,b). Glyphosate formulations available for retail sale are expensive if they must be applied in concentrated form (to \$100/gallon, depending on dilution and brand). IPAW (2007) emphasizes that stems must be freshly, carefully and cleanly cut, that all surfaces must be treated, and that runoff must be prevented. This becomes tedious on younger shrubs with numerous small stems. It is difficult to expect this level of diligence from paid work crews working on a large project, not to mention volunteers and private individuals.

The objective of this work was to test an alternative honeysuckle control method which seemed more consistent with shrub biology and the attributes of glyphosate herbicide. It was expected that the method would be less expensive and could be more easily implemented by volunteers and private landowners. It emphasizes the best practice of limiting quantities of herbicide applied. The rationale for this approach is based on the ecophysiology of honeysuckle in forest understories.

Invasive bush honeysuckles in deciduous forests typically leaf out several weeks before the tree canopy (Barnes and Cottam 1974). Harrington *et al.* (1989) have shown for *L. x bella* that early leaf display significantly enhances carbon gain over closed canopy conditions. Early spring photosynthesis is likely to occur in *L. maackii* as well. We speculated that early canopy development could be exploited to limit energy reserves (storage carbohydrates) available for resprouting, and stimulate the formation of a small volume of replacement foliage amenable to glyphosate spraying. Because spring foliage represents the use of stored carbohydrates to support growth, the period immediately following leaf growth is the time at which whole plant carbohydrate reserves in woody species are at a minimum (Kozłowski and Pallardy 1997). Decapitating the shrub after leafing eliminates any photosynthetic benefit from carbohydrates recently invested in foliage. Additionally, storage reserves to resprout are more limited. Sprouting requires several weeks, which allows the tree canopy to close and limit light availability for understory photosynthesis. Herbicide applied to regrowth can be less concentrated because it is applied to foliage, not woody tissue. Although herbicide overspray is a consideration, regenerated foliage has a far smaller volume and a more compact growth form compared to mature plants.

Two experiments were conducted. The first experiment evaluated whether early growing season cutting treatments might indeed be effective, less chemically intensive control techniques. In this experiment, we also evaluated the success of less concentrated glyphosate applications to stems cut in summer. Because early cutting treatments proved promising, a second experiment was conducted to determine if cutting height (base or at 50 cm) affected the level of control, and verify that herbicide treatment (or other secondary treatment) was necessary for spring cut stems in shaded situations (c.f. Luken and Matimiro 1991).

## METHODS AND MATERIALS

Studies were conducted in a ca. 45 year old upland successional forest dominated by *Robinia pseudoacacia*, *Acer negundo*, and *Ulmus* spp. on the Southern Illinois University Edwardsville Campus (38° 47' N, 90° 00' W), in Madison County, southwestern Illinois. The honeysuckle population in the stand was entirely *L. maackii* (as is typical of the region). Experimental plants were 1.0-2.5 m tall, although individuals in the population ranged from seedlings to > 3 m tall. The overhead canopy was closed, with midsummer light levels < 3% of ambient photosynthetically active radiation. The soil on the site is moderately acid Menfro Silt loam, containing ca. 22% clay in the surface horizons (Leeper 2004).

### Experiment I

In 2004, 80 shrubs were assigned at random to one of four treatments:

- 1) All stems cut to 30 cm height at the time of maximum leaf expansion (early April). Regrowth sprayed with glyphosate in first week of July.
- 2) All stems cut to 30 cm height at the time of maximum leaf expansion (early April). Regrowing foliage cut off at mid growing season (first week of July).
- 3) All stems cut to 30 cm height at mid growing season (first week of July). Coat fresh stubs with glyphosate.
- 4) All stems cut to 30 cm height at mid growing season (first week of July). No subsequent treatment.

Roundup® (Monsanto, Inc., St. Louis, MO) (18% glyphosate) was mixed with tap water at ca. 2X recommended label recommendations for foliar application and 4X recommended concentration for stump application. These solutions correspond to 0.9 and 1.8% glyphosate. Foliage and stems were sprayed using a pump bottle until uniformly wet. Stems were cut with an ordinary garden loppers.

Treatment success was evaluated in mid November (135 days later) by photographing shrubs against a calibrated background, and scoring the regenerated foliage in index classes corresponding to the area of 5 typical leaves. A double blind procedure was used to prevent observer bias. Shrubs with no foliage were classified as dead. Foliage production classes were converted to ranks and analyzed by 1-way ANOVA and linear contrasts. Mortality rates were compared using contingency table analysis with Chi-square and Fisher's Exact tests (Zar 1999).

### Experiment II

In experiment I early cutting with treatment of regrowth proved to be the most effective control technique. We hypothesized that adjusting cutting height might further enhance control. In addition, we speculated that secondary treatment of regrowth might not be necessary in shrubs that are cut at ground level and resprout in deep shade. Experiment II employed a factorial design to evaluate how cutting height and subsequent retreatment (foliar glyphosate application/no treatment) affected control. In addition, light availability and initial shrub size were evaluated as potential confounding influences.

In 2005, 100 shrubs 1.0-2.0 m tall from the same locale as above were assigned at random into four treatment combinations in a 2 X 2 factorial: early cutting (as above) at the

stem base vs. cutting at 50 cm height, and 2X herbicide retreatment of foliage (as detailed above) vs. no retreatment. Before treatment the canopies of all shrubs were measured to determine the lengths of the major and minor axes. These were converted to projected canopy cover. In addition, measurements of photosynthetically active radiation (PAR, photon flux density at  $\lambda=400-700$  nm) were made on four clear days in September over each shrub using a 90 cm Canopy Ceptometer (Decagon Devices, Pullman WA). Mean PAR calculated on log transformed measurements were estimated for each shrub. Leaves were harvested, dried at 60 C, and weighed in mid October. Shrubs having no leaves were classified as dead. Treatment effects on foliage production were analyzed using a mixed model incorporating the simple effects height of cutting, herbicide treatment,  $\log_{10}$  canopy cover,  $\log_{10}$  mean PAR, and 2 way interactions between each pair of simple effects. Foliage production was  $\log_{10}$  transformed after adding 1 to stabilize variances. Mortality rates were compared using multiway contingency table analysis and Mantel-Haenszel  $\chi^2$  to compare responses aggregated across strata (Systat 2000). The influence of canopy cover and PAR on responses to herbicide treatment were evaluated by logistic regression as implemented in Systat 10 (Systat 2000).

## RESULTS

### Experiment I

Experimental treatments produced wide differences in the amount of vegetative regrowth and apparent mortality. Values for the regrowth index ranged from 0 to as high as 10, corresponding to about 50 leaves. Apparent mortality rates ranged from 5% in the shrubs cut at midsummer and not treated with herbicide to 75% in the shrubs cut in early summer and treated with herbicide.

ANOVA of the foliage index revealed highly significant differences between treatments ( $F_{3,76} = 10.84$ ,  $P << 0.0001$ ; Fig. 1a). Time of cutting (early vs. midseason) produced large and significant differences in regrowth ( $F_{1,76} = 30.24$ ,  $P << 0.0001$ ). Index values for shrubs cut in the early season averaged the equivalent of five leaves, while shrubs cut later in the season had about four times more foliage (Fig. 1a). The two different herbicide treatment schemes (herbicide applied to regrown foliage vs. cut stubs) produced widely different effects ( $F_{1,76} = 23.87$ ,  $P < 0.0001$ ). Treatment of regrown foliage produced only about 15% of the regrowth seen in the more concentrated stump treatment (Fig. 1a). Mortality rates paralleled rates of regrowth. Sixty-seven percent of plants cut early in the season appeared dead in November, compared to just 10% of the plants cut in midseason (Fisher's Exact Test  $P << 0.0001$ ; Fig. 1b). Herbicide treatments killed a far greater proportion of plants cut early than plants cut later (75 vs. 15%, Fisher's Exact Test  $P << 0.0001$ ; Fig. 1b). Differing followup treatments *within* a given cutting regime had no effect. For shrubs cut in spring the regrowth indices and mortality did not differ significantly whether shrubs were subsequently sprayed or recut (regrowth index: Fig. 1a,  $F_{1,76} = 1.89$ ,  $P = 0.1729$ ; mortality: Fig. 1b, Fisher's Exact  $P = 0.5006$ ). Shrubs cut in midseason did not respond to herbicide treatment. The regrowth index for herbicide/no herbicide was ca. 20 leaves ( $F_{1,76} = 0.38$ ,  $P = 0.5385$ , Fig. 1a); mortality was about 10% (Fisher's Exact  $P = 0.6049$ , Fig. 1b).

## Experiment II

Manipulations caused large differences in end of season leaf biomass and apparent mortality. Leaf biomass ranged from 0 to 8.2 g; mortality was lowest (4%) in shrubs cut at the base and not treated with herbicide and greatest (85%) in shrubs cut at the base and treated with herbicide. Median shrub canopy cover was 3.8 m<sup>2</sup>, but ranged widely, 0.2 to 20.9 m<sup>2</sup>. Median average PAR for individual shrubs was 30  $\mu\text{mol m}^{-2} \text{s}^{-1}$ , ranging 3-1159  $\mu\text{mol m}^{-2} \text{s}^{-1}$ .

ANCOVA of log<sub>10</sub> leaf mass produced unambiguous results, identifying herbicide application as the single statistically significant determinant of regrowth (Table 1). Least squares means, controlling for all other main effects and the interactions named above, show that the average shrub receiving herbicide treatment produced only 0.33 g leaf tissue compared to 1.74 g produced by untreated shrubs (Fig. 2a). Canopy cover had a statistically ambiguous positive influence on regrowth (Table 1). Notably, light availability and shrub canopy cover did not modify treatment responses: all interactions are nonsignificant (Table 1).

Contingency table analysis showed that cutting height had no effect on mortality (Mantel-Haenszel  $\chi^2 = 0.10$ ,  $P = 0.7500$ ; Fig. 2b). Herbicide application had an important effect, killing 76% of treated shrubs as compared to 8% mortality in untreated shrubs (Mantel-Haenszel  $\chi^2 = 44.01$ ,  $P << 0.0001$ ; Fig. 2b). There were no interactions between cutting height and herbicide treatment (Mantel-Haenszel  $\chi^2 = 0.12$ ,  $P = 0.8888$ ; Fig. 2b).

A limited logistic regression analysis was conducted to detect effects of canopy cover and light availability on responses to herbicide treatment. Neither canopy cover nor light availability had significant effects by themselves, or in interaction with herbicide treatment. Herbicide treatment was significant in both models (Table 2).

## DISCUSSION

Early spring cutting and spraying of regrowth with 0.9% glyphosate (the “new technique”) produced results comparable to reported results for midseason cutting and stump treatment with 20% glyphosate (Kline 1981). For the new technique the mortality rate over two seasons was 80.4% (s.e. = 5.9%). Kline (1981) killed 89.0% (estimated s.e. = 2.7% based on sample size) of shrubs using a 1:5 glyphosate dilution, which might slightly exceed the new technique ( $P = 0.0681$ ). The new technique has the advantage of requiring less herbicide. We estimate that spraying requires about double the volume of herbicide solution, but at 1/10th the concentration is a four fold reduction in herbicide use.

In the first experiment regrowth after early cutting was subsequently sprayed or cut off. On a statistical basis, both secondary treatments had indistinguishable effects, indicating that recutting may be a practical alternative to herbicide. Luken and Mattimiro (1991) suggested that repeated cutting of regrowth showed potential as a control technique in shaded sites. In their study it required four years of cutting in midseason to achieve significant control. Early cutting, as executed here, followed by a few cycles of cutting regrowth may be more effective than Luken and Mattimiro’s approach. Treatment schemes that avoid herbicide application are particularly desirable. Although this project

was undertaken in a very low grade forest with a flora of no particular conservation value, most managers are apt to practice triage, and focus honeysuckle extermination efforts on high quality forests where richer floras are at risk. Recent concerns over losses of herpetofauna and the effects of Roundup® on amphibians (Relyea 2005a,b) should also motivate additional investigations.

The fact that both herbicide application or cutting of regrowth are effective on shrubs cut in early spring supports the proposal that the seasonal distribution of carbohydrates is a major vulnerability in honeysuckle. Based on the results of experiment I, we speculated that one carefully scheduled cutting alone might kill many shrubs by imposing a limited pool of carbohydrate reserves after leaf flush, and the loss of photosynthetic opportunity (lost leaf surface and reduced light availability with canopy closure). Experiment II showed that this speculation was incorrect. Spring cutting without subsequent treatment killed just 12.5% of shrubs. This does not differ meaningfully from 5% mortality after midseason cutting and no secondary treatment in experiment I.

Different cutting heights were evaluated because honeysuckle resprouts from dormant buds on the stems and root crown. It was suspected that cutting at 50 cm would stimulate more abundant regrowth because more dormant buds remained. More extensive regrowth would deplete storage carbohydrates more thoroughly, and provide greater surface for herbicide absorption. Cutting height had absolutely no effect on regrowth or mortality of either herbicide treated or untreated shrubs. From the standpoint of best practices, cutting at the base is preferable because herbicide overspray is apt to be less.

We found limited indication that shrub size, measured by canopy spread, affected treatment response. The ANCOVA of leaf biomass showed a weak ( $P = 0.0860$ ) response to  $\log_{10}$  canopy cover, but the interaction between herbicide application and canopy cover did not influence mortality (Table 2). A study with larger sample size and greater statistical power would likely show that larger shrubs do not succumb as easily as small shrubs. It is not clear whether size might affect treatment success for some physiological reason, or merely because secondary treatment was less uniform on a larger shrub.

Light availability expressed as  $\log_{10}$  PAR had no effect on response. This finding must be interpreted in the context of the wide range of light regimes inhabited by honeysuckle, and the narrower range studied here. Shrubs in the forest interior are significantly light limited to varying degrees. Often honeysuckle inhabits open habitats in which light limitations are far less pronounced. In the shade, regrown shrubs always encountered a significant, albeit probably variable, limitation on photosynthesis. It is a reasonable hypothesis that early cutting and herbicide treatment would be less effective in brighter habitats because new foliage could achieve sustained periods of high photosynthetic rate and support regrowth more easily.

This study provided evidence that early cutting and foliar herbicide application is an effective means to control bush honeysuckles in forest understories. It has the advantage of greatly reducing the total quantity of herbicide applied, but does not produce better results than the traditional approach. Notably, these results refute the assertion that treatment in spring is ineffective. Rather, herbicide application *to stumps* may be ineffective in spring.

Spring treatment has advantages over other times of the year. Early leaf display makes honeysuckle shrubs visible against a background of dormant native species. Cutting also removes the competitive effect of the canopy before the growing season, and stimulates regrowth that is a visible, compact, and physiologically active target for herbicide action. For the private landowner early cutting and retreatment is desirable because herbicide application is less painstaking. Small quantities of properly diluted glyphosate can be purchased in spray bottles for easy application. Moreover, work conditions are more temperate early in the growing season. In forests with significant summer green flora, foot traffic and herbicide spraying need to be carefully controlled when regrowth is treated.

Both the new and traditional methods are considerably more labor-intensive, which can become costly. The traditional method could be the only choice if work crews are otherwise occupied during spring. Invasives campaigns often take place in fall and winter because crews are available. If it is possible to assemble a sufficient number of laborers, the new method proposed here is certainly effective, and expands the time window for restoration work.

### SUMMARY

Asiatic shrub honeysuckles (*Lonicera* spp.) are aggressive invaders of forest and forest edge habitats throughout the Midwest. Control methods for these species are not widely described or evaluated in the open literature. Kline's (1981) method of stem cutting followed by application of 20% glyphosate to the stumps remains widely recommended despite the fact that glyphosate is most effective as a foliar spray as opposed to a stump treatment. We developed and evaluated a cutting and herbicide technique that capitalizes on the normal flux of storage carbohydrates in woody understory plants. For forest understory honeysuckles, stem cutting in spring just after leaf expansion removes foliage before meaningful levels of photosynthesis occur, and probably inflicts a significant cost on the shrubs carbohydrate reserves. Retreatment of regrowth about eight weeks later, either by recutting or spraying with modest concentrations of glyphosate, killed about 80% of treated shrubs. Applications of an approximately equivalent amount of glyphosate to cut stems was ineffective. The technique described here is simpler and cheaper for small organizations and private landowners to implement.

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Table 1. ANCOVA of leaf mass (transformed as  $\log_{10}(X + 1)$ ) remaining at the end of the growing season after manipulations of cutting height and herbicide followup treatment. PAR (photosynthetically active radiation) and projected canopy area were evaluated as influences on treatment response.

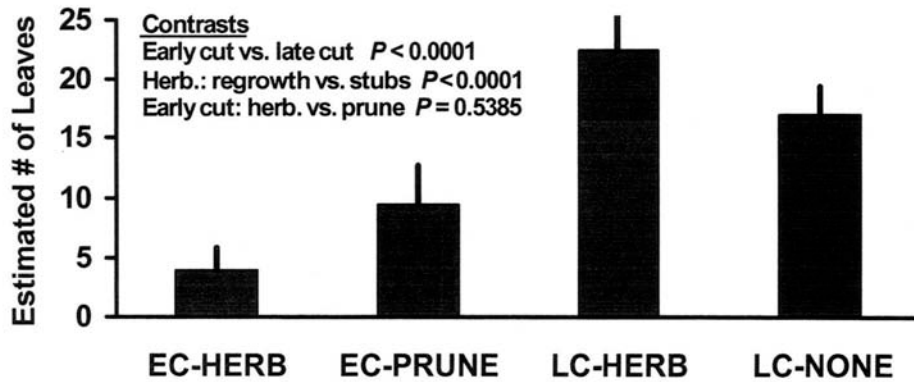
Source	Sum of Squares	Df	Mean Square	F	P
Cutting height	0.1136	1	0.1136	0.06	0.8010
Herbicide	7.9933	1	7.9933	4.50	0.0371
$\log_{10}$ (PAR)	0.7639	1	0.7639	0.43	0.5139
$\log_{10}$ (canopy area)	3.8488	1	3.8488	2.17	0.1451
Cutting height X herbicide	1.1403	1	1.1403	0.64	0.4254
Cutting height X $\log_{10}$ (PAR)	0.7165	1	0.7165	0.40	0.5272
Herbicide X $\log_{10}$ (PAR)	1.5153	1	1.5153	0.85	0.3585
Cutting height* $\log_{10}$ (canopy cover)	0.1228	1	0.1228	0.07	0.7933
Herbicide* $\log_{10}$ (canopy cover)	3.5318	1	3.5318	1.99	0.1625
Residual	134.9418	76	1.77555		

Table 2. Logistic regression of mortality rate as affected by followup herbicide treatment, photosynthetically active radiation (PAR), and projected canopy area.

Estimate	Estimate	S.E.	t	P	Odds ratio
Cutting height	-2.28	0.53	-4.34	0.0000	
Herbicide application	5.69	2.70	2.11	0.0351	296.8
Herbicide application X $\log_{10}$ (canopy cover)	-1.38	1.46	-0.95	0.3445	0.3
Herbicide application X $\log_{10}$ (PAR)	-0.21	1.41	-0.15	0.8802	0.8
LLratio for full model = 67.098147, df = 3, P < 0.0001					

Figure 1 a, b. Responses to spring and fall stem cutting with followup treatments. a. Estimated numbers of leaves based on leaf abundance index ( $\bar{x}$  and s.e.). b. Apparent rate of mortality (proportion and s.e.). Treatment identifiers: EC-HERB, cut early April/herbicide spray early July; EC-PRUNE, cut early April/prune off regrowth early July; LC-HERB, cut early July, spray stumps; LC-NONE, cut early July, no other treatment.

1a.



1b.

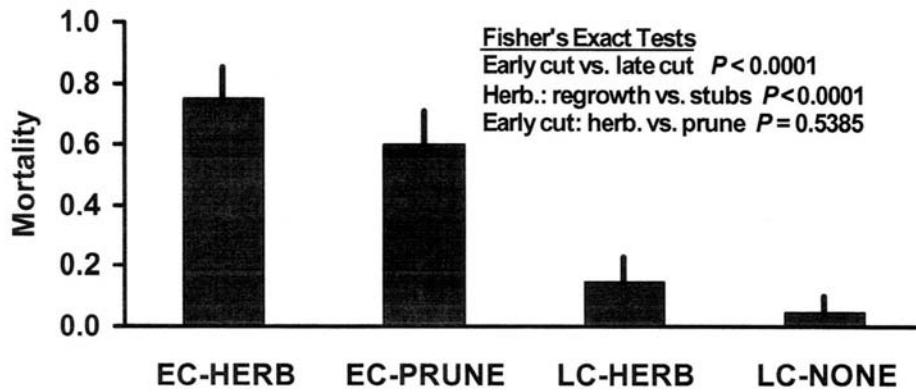
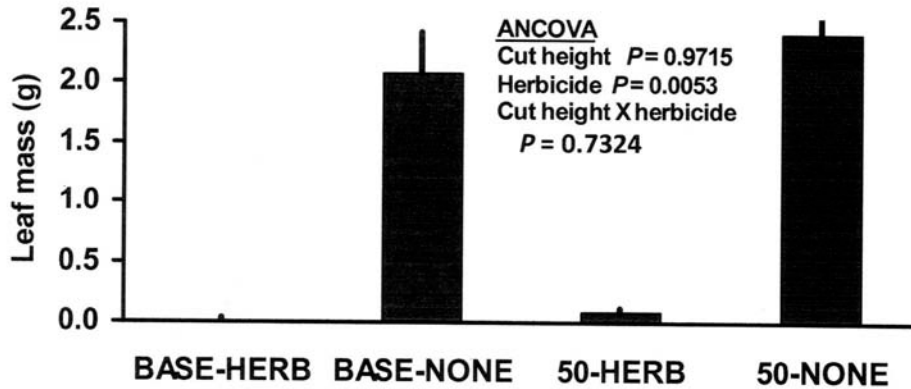


Figure 2 a, b. Responses to height of spring (early April) spring stem cutting and herbicide treatment. a. Dry weight of leaves in October ( $\bar{x}$  and s.e.). b. Apparent rate of mortality (proportion and s.e.). Treatment identifiers: BASE-HERB, cut at base/herbicide spray regrowth early July; BASE-NONE, cut at base, no other treatment; 50-HERB, cut at 50 cm, spray regrowth early July; 50-NONE, cut at 50 cm, no other treatment.

2a.



2b.

