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Targeting Vulnerable Life-Stages of Sericea Lespedeza (Lespedea cuneata) with Prescribed Burns

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There is growing interest in whether invasive species may be controlled by utilizing management strategies that target vulnerable life stages. We manipulated the timing of fire and measured its effects on sericea lespedeza germination and seedling survival. Although fire strongly decreased germination in the laboratory, fire increased germination under field conditions. Additionally, fire caused small decreases in seedling survival in the field. Therefore, controlled burns are likely to encourage spread of sericea lespedeza and are unlikely to effectively control this invasive species. Although targeting vulnerable life stages is a promising strategy for invasive species control, our results illustrate that system-specific studies may be needed to unravel potentially complex interactions between biotic and abiotic factors before effective control strategies can be devised.

Nomenclature: Sericea lespedeza, Lespedea cuneata (Dum. Cours.) G. Don.

Key words: Timing of burn, germination, survivorship.

Invaders possess unique characteristics that allow successful invasion into native ecosystems. For example, one invader may produce a large number of seeds while others may exhibit high growth rates or resource allocation patterns that enhance survivorship in comparison to native species (Milbau et al. 2003; Smith and Knapp 2001). Although the search for the traits that predict invasion has proven difficult, it may be more useful to focus on life stages at which invaders may be most vulnerable to mortality (Ramula et al. 2008). For example, several studies report that the transition from seed to seedling or seedling to adult is critical to population spread for plant species across different ecosystems (Houseman and Gross 2011; Karban and Thaler 1999; Matías et al. 2011; Severns 2003). Presumably, if vulnerable life stages could be identified, control measures could be devised that limit invaders while minimizing impacts of the management strategy on native species and ecosystems (Gurevitch et al. 2011; Schutzenhofer and Knight 2007). For example in prairies, a disturbance regime such as fire could be timed to inflict maximum mortality on encroaching woody species at the seedling or juvenile stage while simultaneously increasing desirable grasses and forbs. Currently, there are few examples of such targeted management except for those utilizing biological control agents (Dauer et al, 2012; Raghu et al., 2006). In this study, we examine the possibility of using fire, a common management tool in grasslands, to disrupt the spread of an invader by targeting its potentially vulnerable life stages.

Sericea lespedeza [Lespedea cuneata (Dum. Cours.) G. Don.] is an herbaceous, long-lived perennial legume initially introduced as a forage crop to the United States from Eastern Asia in 1896 (Cummings et al. 2007). It has since spread throughout the Eastern United States partly from intentional use as a pasture crop and erosion control, and also unintentional spread because of its high propagule production, strong competitive ability, and tolerance of a wide range of environmental conditions (Wei et al. 2009). Sericea lespedeza has profoundly impacted grasslands in the Great Plains by suppressing native plant species. A study in the Chautauqua Hills, Kansas showed that, within 5 to 7 yr of invasion, sericea lespedeza reduced the biomass of native forbs and grasses by 92% and native species richness from 27 to 8 species (Eddy and Moore 1998).

Over the past decade, land managers have attempted to control the spread of sericea lespedeza by using various methods including herbicides, grazing, and fire. A single application of herbicide is largely ineffectual against sericea lespedeza over the long term because, although mature plants can be eliminated, high seed production (up to 6000

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seeds plant^{-1} year^{-1}) creates an extensive seed bank from which new plants can develop (Woods et al. 2009). Consequently, repeated herbicide application is necessary to control and limit spread of sericea lespedeza. Fire does not appear to be a useful tool for controlling mature stands of sericea lespedeza (Munger 2004; Ohlenbusch 2007). In one study, a spring burn once every 3 yr failed to control sericea lespedeza after 6 yr and resulted in large increases in sericea lespedeza cover compared to patch-burned treatments (Cummings et al. 2007).

Little is known about how typical disturbance regimes influence the demographics of sericea lespedeza. Although fire is a common management tool in grasslands that typically suppresses nonnative species and favors native species that are adapted to prairie fires (Heslinga and Grese 2010; MacDonald et al. 2007; Simmons et al. 2007; Smith and Knapp 1999; Suding and Gross 2006), it does not control sericea lespedeza. Fire may be an ineffectual control strategy because adult plants resprout following fire (Munger 2004; Ohlenbusch 2007). However, fire may effectively kill young plants if applied before they have sufficient root reserves to resprout. If this is the case, land managers could potentially control adult plants with herbicide while controlling subsequent recolonization from the seedbank with the application of fire during seedling development.

In addition to potential impacts on immature plants, the effect of fire on germination rates is also unclear. Anecdotal information suggests that fire may stimulate sericea lespedeza germination (Ohlenbusch 2007). However, Bell and Koerner (2009) found that sericea lespedeza seed viability drops significantly at 225 C and seeds are inviable at 250 C. Martin et al. (1975) report that 4 min of exposure to moist or dry heat between 90 and 110 C is lethal to sericea lespedeza seeds. With prairie fires burning at up to 411 C (Engle et al. 1993), sericea lespedeza seed may be vulnerable to fire, but effects of fire on sericea lespedeza seed have not yet been quantified under field conditions.

Prescribed burning has the potential to kill a large number of sericea lespedeza seedlings and consume enough seeds to reduce the likelihood of population spread because of fire. However, the efficacy of prescribed burning is unknown because of the lack of data from combined field and laboratory studies to determine if there are vulnerable life stages and to resolve the apparent discrepancy between anecdotal and laboratory results. In this study, we addressed the following questions: (1) Does fire have suppressive or facilitative effects on sericea lespedeza recruitment? (2) Is this suppressive or facilitative effect on sericea lespedeza recruitment caused by changes in seed germination or seedling survivorship? (3) Is there a plant size or life stage at which sericea lespedeza becomes insensitive to burning within its first two growing seasons? To answer these questions, we manipulated the timing of prescribed burns in a tallgrass prairie to quantify the probability of mortality of sericea lespedeza plants of different ages under field conditions. In addition, we quantified the germination rates of sericea lespedeza seeds under different burning conditions in the laboratory.

Materials and Methods

In order to test the effect of fire on sericea lespedeza at different life stages, two field experiments were conducted at the University of Kansas Field Station (KUFS; 39.05°N, 95.19°W). These experiments were located in a former agricultural field that had undergone natural conversion to tallgrass prairie since 1956.

Field Experiment 1. We tested whether burning in the fall shortly after seed dispersal would reduce recruitment of sericea lespedeza. Since sericea lespedeza disperses seeds in fall and viable seeds do not typically remain on stems, it was hypothesized that a spring burn may be ineffectual if seeds become incorporated into the soil over the winter because of freeze–thaw action. Consequently, a fall burn, after dispersal but before seeds become incorporated into the soil might have large, negative effects on the viability of recently dispersed seeds. However, if a fall burn had weak effects on seed mortality, germination might increase because of removal of litter and changes in soil conditions. To test whether a fall burn would have negative effects on the viability of recently dispersed seeds, two treatments (seed addition either pre- or post- burn), were applied to 16, 0.5 by 0.5-m (1.64 by 1.64 ft) plots in a complete block design with eight replicates of each treatment. Seeds harvested from near the study site were sown on the soil surface at 150 seeds per plot with the number of seeds estimated from a seed mass–number relationship. For the
preburn treatment, seeds were sown by hand on October 29, 2009. The field burn was conducted on November 5, 2009. After the burn, on November 6, 2009, the postburn seeds were sown by hand and all plots were uniformly covered with erosion control fabric until March 18, 2010 to prevent the potential movement of seed by wind or heavy rain. Plant density was surveyed after two growing seasons on September 10, 2011. A one-way, blocked ANOVA was performed in SAS 9.2 (SAS Institute Inc., Cary, NC, USA) to determine the effects of pre- and post-burn seed additions on sericea lespedeza density after two growing seasons.

**Field Experiment 2.** A second experiment at KUFS was initiated to test the effects of fire on sericea lespedeza germination and survivorship of seedlings of different ages. On March 18 2010, 90, 1-m by 1-m plots were arrayed in a grid with 0.5 m between plots. Six burn and seed addition treatments were applied in a randomized complete block design that was replicated ten times. Sericea lespedeza seed, from the same source used for Experiment 1, was sown by hand on the soil surface of each plot. The seeding rate was 4000 seeds per plot with seed numbers estimated from a seed mass–number relationship. Each block contained one plot burned on April 21, May 25, June 21, July 21, and September 4 of the first growing season (2010) and April 21 of the second growing season (2011). A tallgrass prairie burn was simulated in each plot using a propane torch to ignite the vegetation. Temperature was monitored using temperature-sensitive paint (Tempil, South Plainfield, NJ, USA) applied to aluminum strips that were placed within the plot prior to the burn. This allowed us to ensure that the burns were within the 350 to 400°C range typical of prairie burns (Gibson et al. 1990; Hobbs et al. 1991; Middleton 2002; Engle et al. 1993).

In order to relate seedling age to survivorship, every 20 d throughout the growing season, up to four sericea lespedeza seedlings in each plot were randomly selected and marked by placing a metal ring around each seedling. All unmarked seedlings in each plot were removed every 10 d to ensure that seedlings in the next cohort were no more than 10 d old at the time they were marked. In total, six cohorts were marked in each plot over the course of the experiment with the first cohort being marked on June 2, 2010. First-year survivorship of these cohorts was quantified on November 4, 2010 and survivorship at the end of the second growing season was recorded on August 2, 2011. In the event that a ring was not found, its corresponding data point was dropped from the analysis. Cumulative germination was quantified based on the sum of marked and removed seedlings from each plot in the first growing season and April 21 of the second growing season. Since sericea lespedeza do not set seed until the end of their second growing season, all seedlings within experimental plots germinated from the sown seed.

To test the effect of burn timing on the survival of sericea lespedeza seedlings, we used linear regressions with plant age at the time of burn (plant age) and the timing of burn as independent variables and percent survivorship within each plot as the response variable. Here, timing of burn refers to the number of growing days elapsed between the initial emergence of sericea seedlings in the spring of the first growing season and the day of each of the burn treatments. Growing days were limited to days between April and November. Differences in cumulative germination between burn treatments were tested with a one-way ANOVA for a randomized complete block design using SAS 9.2 (SAS Institute Inc., Cary, NC, USA). Bonferroni correction was utilized to control for multiple comparisons. Unless otherwise stated, statistical significance was assessed at $P \leq 0.05$.

Survivorship was predicted to increase with plant age, as older or larger plants are more likely to have sufficient root reserves to resprout following a burn than younger plants. To relate survivorship to plant size of the various cohorts utilized in the field experiment, we conducted a greenhouse experiment to determine the strength of the size–age relationship. Thirty-two intact plant communities and the associated soil (approximately 21-cm-diam [8.27 in], 19-cm deep) were removed from an area adjacent to the Field Experiment 2 plots. These plant communities were transferred to cylindrical pots (diam = 21.5 cm, ht = 21.5 cm) and randomly arranged on a greenhouse bench. Five seeds of sericea were sown into each pot. To insure maximum germination, seeds were scarified with sulphuric acid prior to sowing (Bentley, 1933) and watered once every 2 d. Every 20 d, sericea seedlings from four randomly selected pots were harvested by saturating soils with water and carefully removing sericea lespedeza plants. These harvests corresponded to the burn intervals utilized in Field Experiment 2. Harvested plants were dried for 48 h at 60 C (140 F) and root and shoot length and dry weight were measured. Simple linear regressions of shoot and root length and mass on plant age were performed in SAS 9.2 (SAS Institute Inc., Cary, NC, USA).

**Lab Germination Experiment.** A laboratory burn experiment was conducted to test how fire, light, and litter influenced sericea lespedeza germination rates. After a complete burn, photosynthetic photon flux density (PPFD) is at or near 100% at the soil surface, but under unburned conditions, standing plant material and litter reduces light that reaches the seeds and the soil, potentially altering germination rates (Baskin and Baskin, 2000). The presence of litter may also affect the amount of heat reaching the seeds during the burn, potentially preventing them from being consumed by the fire, or by altering soil chemistry (Blank and Young 1998; Keeley and Bond 1997). Such changes in light, temperature and nutrients are likely to contribute to germination but the magnitude and direction
of these effects are difficult to predict (Baskin and Baskin, 2000).

Four burn treatments were devised to separate the direct and indirect effects of burning on sericea lespedeza germination: (1) unburned seed was added to unburned soil with unburned litter as a control for burning; (2) unburned seed was added to burned soil and burned litter to test whether physical or chemical changes to the litter or soil affected germination; (3) burned seed was added to unburned soil and unburned litter to test if burning alters germination regardless of fire-induced changes to soil and litter; (4) and soil, litter, and seed were all burned to test their combined effects on germination. To examine the potential interaction between light and burning on germination, we manipulated light availability with either 60% shade cloth to reduce PPFD to 40% or an unshaded control, mimicking light levels measured in the field (38% or 100% PPFD, respectively). Burning and light treatments were combined factorially and replicated 12 times in a completely randomized design.

One hundred sericea lespedeza seeds were used in each treatment. Soil was collected from a site adjacent to the field experiments and sieved before treatments were applied. All burns were conducted using a propane torch (Bernzomatic, Medina, NY, USA) in an aluminum tray and mimicked the temperature of simulated field burns (350 to 400 C), which were monitored with temperature-sensitive paint (Tempil, South Plainfield, NJ, USA) applied to aluminum strips. Since the duration of fire is typically short in prairie burns, the flame was evenly passed over the tray for approximately 30 s per treatment. After the burn treatment, the soil was transferred to 25-cm by 25-cm by 4-cm plastic trays which were kept below 0 C for 2 wk to simulate overwintering. Each tray was covered with a transparent cover to reduce desiccation and the assigned shade cloth treatment, and placed on an illuminated shelf. Trays were then watered daily and germination was quantified over 17 d.

The effects of light and burn treatments on the germination rate were tested using a two-factor ANOVA in SAS 9.2 (SAS Institute Inc., Cary, NC, USA) with Bonferroni correction for multiple comparisons.

Results and Discussion

Fire and Survivorship. Field Experiment 1. The effect of fall burning had a large effect on the establishment of sericea lespedeza after two growing seasons ($F_{1, 7} = 17.39$, $P = 0.0042$). Plant density (mean ± SE) was higher when seeds were sown postburn (7.25 ± 1.1, $n = 8$) than preburn (1.63 ± 0.6, $n = 8$).

Field Experiment 2. Simple linear regression revealed that, although seedling survivorship was significantly influenced by plant age ($t_{1, 151} = 3.56$, $P < 0.0001$) and the timing of burns ($t_{1, 151} = 2.59$, $P = 0.010$), these factors explained very little of the variation in seedling survivorship ($r^2 = 0.071$ and $r^2 = 0.036$, respectively). As we predicted, vulnerability to burns decreased as plants aged but this effect was weak relative to the other factors that influenced seedling survivorship. Such variability may be driven by resource competition, herbivory, pathogens, spatial variation or high genetic variability in seed quality. Furthermore, evidence from the greenhouse growth rate experiment revealed that plant age is weakly correlated with root ($r^2 = 0.170$) and shoot ($r^2 = 0.176$) mass, which suggests that, although plant size does increase with plant age, there is a high amount of variability in plant size during the first growing season because of factors other than plant age even under controlled conditions. This variation in size among plants is likely to have strong effects on their potential to resprout following a burn and makes it difficult to determine an approximate age at which seedlings become invulnerable to fire.

The application and timing of prescribed burns influenced the survivorship of sericea lespedeza. Mean seedling survivorship was higher in unburned control plots than in any plot that experienced a burn. Plants that germinated preburn had increased mortality compared to those that germinated postburn or in unburned controls (Figure 1). However, the advantages to the seedling of germinating post-burn were reduced as the timing of the burn approached the end of the growing season. For

Figure 1. The effect of the burn date, regardless of plant age, on sericea lespedeza seedling survivorship in the field. Sericea lespedeza survivorship (mean ± 1 standard error) at the end of the second growing season (August 2011) for seedlings that germinated before a burn (black) or after a burn (gray). Seedlings that germinated prior to burning had lower survivorship than those that germinated after a burn or those in unburned controls.
example, there was extremely low survivorship among plants that germinated after the September burn even though sericea lespedeza continued to germinate through November 3, 2010, which suggests that, despite ongoing germination throughout the year, any seedlings germinating after September will not survive the winter.

**Fire and Germination. Field Experiment 2.** We found that burning had strong, positive effects on cumulative germination in Field Experiment 2 through April of the second growing season ($F_{6, 54} = 6.27, P < 0.0001$). Cumulative germination in plots burned in April, May, June, July, and September of the first growing season were similar and higher than the unburned control (Figure 2).

Although there are anecdotal reports that fire stimulates sericea lespedeza germination, (Ohlenbusch 2007) there are little or no data from field experiments to quantify the effect of fire on sericea lespedeza germination. Results from Field Experiment 2 suggest that burning does have a direct stimulatory effect on germination since every burn treatment increased sericea lespedeza germination approximately twofold by the second growing season regardless of when the burn occurred (Figure 2). In addition, there was a flush of germination in the first growing season directly following each burn event regardless of the timing of the burn during the growing season (Wong, 2011). The stimulatory effect of fire on germination in the field could be a result of scarification of the seeds (Herranz 1998), heat shock breaking dormancy (Herranz 1998; Gashaw and Michelson 2002; Baiza and Vallejo 2005), or perhaps the reduction in competition for resources because of the removal of aboveground biomass (Potvin, 1993).

**Laboratory Germination Experiment.** In order to test whether the stimulatory effects of fire in the field were because of direct effects on seeds or indirect effects on resources, we tested for the effects of light, fire, and soil conditions on germination in the laboratory. An experimental reduction from 100% to 40% light availability had no detectable effect on sericea lespedeza germination rate ($F_{1, 77} = 0.80, P = 0.37$). However, the various burn treatments had large, negative effects on germination rates ($F_{3, 77} = 423, P < 0.0001$; Figure 3). Germination rates of unburned seeds added to burned soil and litter did not significantly differ from the unburned control suggesting that changes in soil chemistry because of fire do not affect sericea lespedeza germination. In contrast, when soil, litter, and seed were burned, germination rates were 68% lower than unburned controls indicating that fire can inflict substantial mortality on sericea lespedeza seeds (Figure 3). When seeds were burned but litter and soil remained unburned, germination was lower than all other treatments.

Effects of burning on sericea lespedeza germination differed between the field and laboratory experiments illustrating that complex interactions occurring under field conditions may not be revealed in lab experiments.

![Figure 2. Effect of burn timing on the cumulative germination rates (mean ± 1 standard error) of sericea lespedeza seedlings in the field by April of the second growing season grouped by burn treatment. Plots subjected to any burn treatment resulted in higher cumulative germination by the second growing season compared to unburned controls.](image)

![Figure 3. Effect of burning seed, soil, and litter on sericea lespedeza germination (mean ± 1 standard error) in the laboratory under 100% (gray) and 40% (black) light availability. Light availability had no effect on germination. Burning seed substantially reduced germination compared to unburned or burned litter/soil. Seed burned on soil with litter had higher germination rates than seed burned alone. Different letters indicate significant differences among burn treatments irrespective of light availability.](image)
potentially leading to contradictory conclusions. In the Lab Germination Experiment (Figure 3), there was an extremely large negative effect of fire on sericea lespedeza germination while, in Field Experiment 2, cumulative germination in burned plots exceeded the unburned controls. Although we controlled burning duration and intensity in the field and laboratory, differences in burning conditions may have altered the intensity of the fire in the laboratory compared to the field, which could lead to a larger negative effect on the seed germination. A more likely explanation for these contradictory results is that seeds became incorporated into the soil through heavy rains and freeze–thaw events that may have protected seeds from the burns. Although our experiments did not fully tease apart the stimulatory effects of fire on germination, they did illustrate that laboratory germination experiments without the associated field experiments could lead to erroneous management recommendations as fire may affect germination to a greater extent in the field than in the laboratory.

Our germination results in the field are generally consistent with those found in the literature. The higher cumulative germination in Field Experiment 2 is consistent with legumes subjected to field burns in dry Mediterranean valleys (Baeza and Vallejo 2006). The low germination of burned seeds and the high germination of unburned seeds in our laboratory experiment is consistent with one study (Vermeire and Rinella 2009) but contrasts what is commonly reported in laboratory studies where burning breaks the seed coat and enhances germination (Blank and Young 1998; Gashaw and Michelson 2002; Keeley and Bond 1997; Keeley 1987). Additionally, heat-treated seeds have enhanced germination in some laboratory experiments (Martin et al. 1975; Qiu et al. 1995; Segelquist 1971); however, these studies did not use fire nor did they use temperatures as high as those used in our experiments. Furthermore, in our experiment, the unburned seeds sown into trays with burned soil and litter were presumably exposed to chemical changes of burned soil and litter, but this exposure did not alter germination as other studies would suggest (Blank and Young 1998; Keeley and Bond 1997). Additionally, the fact that burning has strong negative effects in the laboratory but mixed to positive effects in the field illustrates how complexities of these interactions can be obscured when using only laboratory assays.

We hypothesized that targeting sericea lespedeza with fire at a young life stage could be an effective management strategy particularly by controlling reestablishment from the seed bank. Results from Field Experiments 1 and 2 provide support for this strategy as survivorship of sericea lespedeza seedlings was reduced by fire (Figure 1). However, this decrease in survivorship was mitigated by the enhanced germination in the field so that the net effect of burning is unlikely to reduce establishment of plants from seed.

This study is not without its limitations including both time and space. The study was conducted over the course of two growing seasons and the results are subject to the conditions of those particular years. It should also be noted that the experiment was conducted at a single site and variation in soils and climate may alter germination and growth rates. Likewise, this experiment necessarily included practical limitations such as the use of simulated burns rather than field-scale burns. Despite these limitations, our results illustrate how fire can interact with environmental variables at early life stages of sericea lespedeza to influence the performance of this invader in grassland habitats. Furthermore, the results illustrate that, although targeting vulnerable life stages may be a good strategy to control the spread of invaders, additional studies are needed to insure that unforeseen interactions do not lead to ineffectual or detrimental management outcomes.

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