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Effects of leaf litter and precipitation on germination and seedling survival of the endangered tree *Beilschmiedia miersii*

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Abstract

Question: What effects do leaf litter and rainfall regime have on seed germination (time and probability) and seedling survival of the endangered tree species *Beilschmiedia miersii* (*Lauraceae*)?
Location: The species is a native tree from the mediterranean climate region of Chile. Seeds were collected from La Campana National Park (Chile). The study was carried out under controlled conditions at the Laboratory of Ecology, University of Chile, Santiago, Chile.
Methods: During April 2001, 200 seeds were assigned to four experimental treatments: high precipitation, with and without litter and low precipitation, with and without litter. Each treatment had 50 individual seeds, each seed in an individual pot. For statistical purposes, we considered each seed as one replicate. High and low values of artificial rainfall corresponded to mean dry and wet years, respectively, for the period 1958 - 1993 in the central zone of Chile.
Results: Seeds germinated earlier, and in higher proportion, in the presence of leaf litter, but only under low rainfall. Seedling survival was insensitive to both litter and precipitation.
Conclusions: We conclude that the presence of litter in native populations facilitates seed germination and recruitment of *B. miersii*, particularly during dry years. We suggest that the reduction of leaf litter due to extraction for gardens and horticultural activities might preclude regeneration of this endangered species.

Keywords: Chile; Facilitation; Mediterranean ecosystem; Plant recruitment; Plant regeneration; Threatened species.

Introduction

Accumulation of leaf litter modifies light, moisture and microhabitat environment; hence it may affect seed germination and seedling survival (Harper 1977; Rathcke & Lacey 1985; Facelli 1994). Leaf litter negatively affects seed germination and seedling growth, e.g., acting as a mechanical barrier for radicular growth (Bosy & Reader 1995) and shoot emergence (Green 1999). Leaf litter may also modify the quantity and quality of light received by a seed in the ground, transmitting longer wavelengths that may be strongly inhibitory for the germination of seeds (Facelli & Pickett 1991a; Yirdaw & Leinonen 2002). Leaf litter may also modify the chemical environment of seeds, inhibiting germination (Bosy & Reader 1995; Hilhorst & Karssen 2000). On the other hand, leaf litter may positively affect seed germination due to its role as refuge against predators (Myster & Pickett 1993; Cintra 1997), or its moderating effect on soil temperature and moisture (Facelli & Pickett 1991a; Bosy & Reader 1995).

The sign and magnitude of litter effects are variable and seem to depend on the regeneration requirements of plant species and on ecosystems properties such as climate, soil and vegetation (Rice 1979; Carson & Peterson 1990; Facelli & Pickett 1991b; Bosy & Reader 1995; Foster & Gross 1997).

Leaf litter effects on soil moisture may be crucial for water demanding species. However, the favourable effect on these species may depend on the amount of rain that falls in a given season or at a particular place (Callaway & Pugnaire 1999). Leaf litter effects on soil moisture are important when rainfall becomes scarce. By reducing evaporation from the soil, leaf litter maintains soil moisture necessary for seed germination during dry periods or at dry sites (Callaway & Pugnaire 1999). This potentially important interaction between leaf litter and rainfall has not yet been experimentally assessed.

In forests of central Chile, leaf litter cover and biomass vary spatially, primarily because of two human caused disturbance mechanisms: reduction of woody plant cover due to logging, thus decreasing leaf litter sources (Rundel 1981) or through direct extraction of leaf litter from forest soils for home gardens and other horticultural purposes (Lienlaf 1996). Precipitation in central Chile is also variable between, and within, years as well as with latitude (Di Castri & Hajek 1976; Santibañez & Uribe 1993). Thus, it is possible to find a gradient of environments where either leaf litter or precipitation varies in space and time. In this study we experimentally examined the effect of leaf litter on seed germination and seedling survival of *Beilschmiedia miersii* (*Lauraceae*), under two contrasting, simulated precipitation regimes in the laboratory.

B. miersii is an endemic and endangered (Benoit 1989) evergreen tree species that occurs in coastal forests in central Chile from 32°33' to 34°02' S (Rodríguez et al. 1983). It is distributed along humid and shaded environments (ravines and south facing slopes) in the mediterranean region of Chile (Armesto & Martínez 1978; Villaseñor & Serey 1980; Donoso 1993). It is a shade-tolerant tree, up to ca. 25 m tall; its fruits are drupes (4 cm long, 2 - 3 cm wide), the largest fruits for all mediterranean trees species of central Chile (Rodríguez et al. 1983), and currently no dispersal agent is known (Henríquez & Simonetti 2001). This species is considered under risk of extinction because of intensive exploitation, habitat destruction and seedling herbivory by cattle (Rundel 1981; Henríquez & Simonetti 2001). La Campana National Park (Biosphere Reserve) (32°47' S, 71°11' W) supports the only protected populations (Henríquez & Simonetti 2001). According to its high soil moisture requirement, we expected that both high rainfall and the presence of leaf litter will have positive effects on seed germination and seedling survival. However, we expected that the effect of leaf litter would be more important as rainfall becomes limiting.

Table 1. Monthly distribution of irrigation (equivalent in mm) supplied to high precipitation (HP) and low precipitation (LP) treatments for the duration of the experiment.

Month	HP	LP
May	63	27
June	112	48
July	121	52
August	67	29
September	41	18
October	15	6
Total	**419**	**180**

Methods

Experimental design

During January 2001 we collected *B. miersii* seeds from one population located on a homogeneous habitat in La Campana National Park, a typical mediterranean climate site. Seeds were obtained from the forest floor during the dispersal period. Immediately after collection, seeds were stratified at 5 °C for 60 d to break induced dormancy (Anselmo 1998). Seeds were planted individually into a 50:50 mixture of forest soil (from La Campana National Park) and sterile sand 1000cm^3 pots. We assigned *B. miersii* seeds to four treatments: (1) high precipitation with leaf litter, (2) high precipitation without leaf litter, (3) low precipitation with leaf litter and (4) low precipitation without leaf litter. Seeds placed in 'with leaf litter' treatments were totally covered with litter. Intact litter was obtained from forest stands in La Campana National Park and was composed of dead leaves from *B. miersii* and other tree species such as *Cryptocarya alba*, *Persea lingue*, *Quillaja saponaria* and *Lithraea caustica*. Litter was homogenized prior to use in the experiments. Each treatment had 50 individual seeds, each in an individual pot. For statistical purposes, we considered each seed as one replicate. High and low precipitation were simulated based on precipitation records from the meteorological station Rinconada de Maipú (33°28' S, 70° 51' W) for the period 1958 - 1993. This station is representative of mediterranean climatic conditions of central Chile (Di Castri & Hajek 1976). We divided years into wetter and drier years (50% each), and calculated the mean for each group. Thus, the irrigated quantity for each precipitation treatment corresponded to the mean value of wetter (419 mm) and drier (180 mm) years. The amount of water to be supplied to seeds each month was calculated by calculating the mean monthly precipitation for dry years and for wet years separately (Table 1). Seeds were irrigated three times per week during winter (June - August) and twice per week during spring (September - November). All assays were conducted in the laboratory at a temperature of 20 °C.

Statistical analysis

We evaluated the following response variables:

1. Germination time: the time elapsed before visible radicule emergence (weeks) since seeds were planted; we conducted a two-way ANOVA for ranked data (Scheirer-Ray-Hare test (*H* test), Sokal & Rolf 1995) to evaluate the effect of leaf litter and precipitation on germination time as this variable did not meet normality assumptions ($X^2_{(0.05,\ 7)} = 117.34$, $P < 0.0001$). The

statistical analysis for this response variable was conducted only with germinated seeds ($n = 94$).

2. Seed germination probability: we assigned 1 if a seed germinated and 0 if it did not.

3. Seedling survival probability: we assigned 1 if a seedling survived and 0 if it did not. To evaluate the effect of leaf litter and irrigation (categorical factors) on seed germination and seedling survival we used a binomial linear model with logit link (see Visual GLIM procedure in StatSoft, Inc. 2001. STATISTICA (data analysis software system), version 6. www.statsoft.com). In this analysis, we considered individual seeds as independent replicates. For seed germination analysis, the sample size was 200 seeds (50 replicates per treatment) i.e. the total number of seed initially used in the experiment; for seedling survival analysis, the sample size decreased to 94 i.e. the number of seeds that effectively germinated, as 106 seeds did not germinate during the experiment.

Results

From 200 seeds initially planted, 47% germinated and 36% survived to seedling stage. Germination time was significantly decreased by the presence of leaf litter (ANOVA for ranked data, $H = 9.65$; df = 1; $P = 0.001$, Fig. 1). In contrast, irrigation treatments did not have significant differences (ANOVA for ranked data, $H = 0.026$; df = 1; $P = 0.87$, Fig. 1). However, we detected a statistically significant interaction between irrigation and leaf litter (ANOVA for ranked data, $H = 4.01$, df = 1; $P = 0.045$), where leaf litter decreased germination time only under the low irrigation treatment (Fig. 1).

The presence of leaf litter (likelihood type 1 test, $X^2_{(0.05, 1)} = 29.74$, $P < 0.0001$) and high irrigation (likelihood type 1 test, $X^2_{(0.05, 1)} = 7.68$, $P = 0.006$) significantly increased the proportion of seeds germinated at the end of experiment (Fig. 2a). However, there was a significant interaction between these two factors (likelihood type 1 test, $X^2_{(0.05, 1)} = 5.79$, $P < 0.02$, Fig. 2a).

Finally, the presence of leaf litter (likelihood type 1 test, $X^2_{(0.05, 1)} = 0.08$, $P = 0.76$) and high irrigation (likelihood type 1 test, $X^2_{(0.05, 1)} = 0.0005$, $P = 0.98$) did not enhance seedling survival (Fig. 2b), and there was no significant interaction between these two factors (likelihood type 1 test, $X^2_{(0.05, 1)} = 0.13$, $P = 0.72$, Fig. 2b).

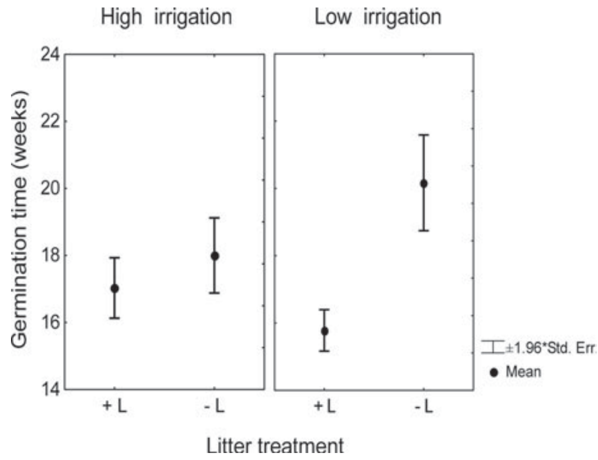

Fig. 1. Germination time of *B. miersii* seeds for different treatments of leaf litter and irrigation under laboratory conditions; + L = with litter; – L = without litter.

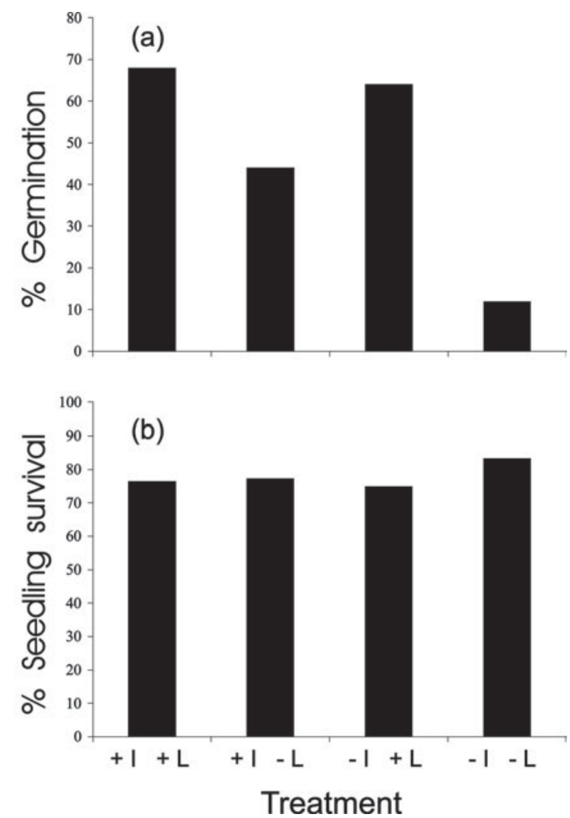

Fig. 2. (**a**) Germination probability and (**b**) Seedling survival of *Beilschmiedia miersii* seeds for different treatments of leaf litter and irrigation under laboratory conditions. + I = high irrigation; – I = low irrigation; + L = with litter; – L = without litter.

Discussion

We showed that: (1) leaf litter significantly decreased germination time under low irrigation conditions, (2) leaf litter increased seed germination percentage under low irrigation and (3) neither leaf litter nor irrigation significantly affected seedling survival. From these results, it is clear that for *Beilschmiedia miersii*, the presence of leaf litter is more important for seed germination than for seedling survival, with this effect being positive, in contrast to most other studies (e.g. Bosy & Reader 1995; Hilhorst & Karssen 2000). Thus, we demonstrate that what is favourable for seeds is neutral for seedlings. Moreover, this result confirms the need to include various transitions during the life cycle of a plant in regeneration studies (Harper 1977).

Results of this study may be better understood by considering particular seed and seedling attributes of this endangered tree species. Seeds of *B. miersii* do not survive from one year to another therefore, any factor (such as the presence of leaf litter) that assures seed access to moisture and precludes early desiccation will enhance seed germination and prevent mortality (Chacón & Bustamante 2001). Interestingly, facilitation effects of litter were important only under low irrigation conditions. This is a result that supports the hypothesis that the importance of leaf litter for plant regeneration is expressed under water stress (Carson & Peterson 1990; Facelli & Pickett 1991a; Bosy & Reader 1995; Jensen & Meyer 2001), and that facilitation interactions, in this case by leaf litter, may occur when environmental stress is higher (Callaway & Pugnaire 1999).

The lack of effects of litter and irrigation on seedling survival may be explained by considering the size of *B. miersii* seeds, among the largest of mediterranean flora (Fuentes et al. 1986). It is possible that the endosperm of such a large seed provides resources to emergent seedlings, making them relatively independent from external hazards (Green 1999; Kitajima & Fenner 2000). Thus, seed reserves would determine a similar seedling survival among treatments. By this mechanism, this species, like other *Lauraceae* species (Foster 1986), has the potential to form a seedling bank (Silvertown & Charlesworth 2001).

B. miersii is considered an endangered tree species (Benoit 1989). At present, 20-30 populations of *B. miersii* are known (Serra et al. 1986; Gajardo et al. 1987), but only two of them are protected (Henríquez & Simonetti 2001). The cause of this endangered status is the intense exploitation for wood in the recent past (Rundel 1981; Serra et al. 1986). Nowadays, the small populations are subjected to additional threats such as habitat destruction and fragmentation (Rundel 1981; Serra et al. 1986; Simonetti & Cornejo 1990), and natural regeneration of this and other tree species are threatened by livestock grazing (Henríquez & Simonetti 2001). In this study, we identify other potential threats to conservation of this species. The reduction of plant cover due to deforestation diminishes and direct leaf litter removal for horticultural purposes (Lienlaf 1996) will reduce germination in this species. This is particularly serious because most *B. miersii* populations are outside protected areas (Henríquez & Simonetti 2001). Further, seeds dispersed in populations where leaf litter has been removed will be more exposed to seed predation by herbivores because removal of litter increases the time to germination (Myster & Pickett 1993; Cintra 1997). This effect may become a serious threat for plant regeneration in protected areas where livestock is allowed to enter without control (Henríquez & Simonetti 2001).

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