# Benefits of Using Shrubs as Nurse Plants for Reforestation in Mediterranean Mountains: A 4-Year Study

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## Abstract

**Shrubs are commonly considered competitors of planted seedlings in reforestation programs. However, shrubs can facilitate the establishment of understory seedlings in environments that, like Mediterranean-type ecosystems, are characterized by harsh environmental conditions. In 1997, an experiment was set up in the Sierra Nevada mountains (southeast Spain) to test the use of shrubs as nurse plants for an alternative reforestation technique. Two-year-old seedlings of *Pinus sylvestris* and *Pinus nigra* were planted in four microhabitats: (1) open interspaces without vegetation (which is the usual method employed in reforestation programs), (2) under individuals of the shrub *Salvia lavandulifolia*, (3) under the north side of spiny shrubs, and (4) under the south side of spiny shrubs. Seedlings were also distributed in plots with and without ungulates to test the effect of herbivore damage. We report here the results of survival and growth after four growing seasons, a time span long enough to draw robust conclusions concerning the suitability of this technique. Pine survival was remarkably higher when planted under individuals of *S. lavandulifolia* as compared with open areas (2.6 times for *P. sylvestris* and 1.8 for *P. nigra*). The survival of both pine species was also higher when planted on the north side of spiny shrubs, while mortality on the south side was similar to that found in open areas. The reduction of solar radiation by the canopy of shrubs was likely the main factor determining shrub facilitation. The growth of the pines differed among years. However, growth was not inhibited when planted with shrubs as compared with open areas in any of the years. Herbivore damage was low but was mostly concentrated in the leader shoot, exacerbating the deleterious effect of ungulate herbivores on pine growth. We conclude that the use of shrubs as nurse plants for reforestation is a viable technique to increase establishment success of reforestation in Mediterranean-type ecosystems and that it might be similarly useful in other water-stressed environments. In addition, this technique offers the advantage of following natural succession, thus minimizing the impact in the community.**

**Key words: facilitation, Mediterranean ecosystems, nurse plants, *Pinus*, silviculture.**

## Introduction

Reforestation is a major environmental goal worldwide (Savill et al. 1997; FAO 2001; Lamb 2001). In areas with a long history of human population, such as the Mediterranean basin, intense exploitation has, in many cases, altered the original composition of species and the physical environment (e.g., erosion) (Bauer 1991; García-Salmerón 1995; Blondel & Aronson 1999). Under such circumstances, planting is particularly necessary to recover the vegetation and to ensure the success of regeneration (Bauer 1991; García-Salmerón 1995; Serrada 1995). However, in the Mediterranean basin, stressful environmental conditions prevail that are not usually suitable for the survival of planted seedlings. Therefore, reforestations often suffer heavy plant losses that hinder restoration efforts, mostly as a consequence of summer drought and ungulate browsing (Mesón & Montoya 1993; García-Salmerón 1995).

The development of suitable strategies for forest regeneration in the Mediterranean basin is an issue of primary concern (García-Salmerón 1995; Scarascia-Mugnozza et al. 2000). In the prevailing reforestation techniques used in the region, the preexisting shrubby vegetation is considered to be a source of competition for the newly planted young trees (Mesón & Montoya 1993; García-Salmerón 1995; Serrada 1995), and consequently, shrubs are cleared by different methods or seedlings are planted far from shrubs (García-Salmerón 1995; Serrada 1995; Sternberg et al. 2001). This approach derives primarily from models developed in central and northern parts of Europe (Groome 1989; Bauer 1991), where dense vegetation cover and the lack of severe drought stress make the preexisting shrubby vegetation a source of competition for nutrients and light, increasing the mortality of the reforested seedlings (Savill et al. 1997). However, interactions among plants may be positive in environments

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that, like Mediterranean-type ecosystems, are characterized by harsh physical conditions such as summer drought accompanied by high temperatures (Bertness & Callaway 1994; Callaway 1995; Brooker & Callaghan 1998). In such settings, the preexisting vegetation may offer protection to seedlings by buffering microclimatic extremes (i.e., reducing solar radiation and soil temperature, conserving soil moisture, and enriching nutrient content) and, in addition, may protect seedlings from herbivore damage (Callaway 1995; Rousset & Lepart 1999; García et al. 2000; Gómez et al. 2001a, 2003). In fact, a growing body of experimental studies is reporting a facilitative effect of shrubs and grasses for the early establishment of reforested woody species in Mediterranean environments (Maestre et al. 2001; Gómez et al. 2001b, in press; Castro et al. 2002).

In 1997, an experiment was set up in the Sierra Nevada mountains (southeast Spain) to test an alternative technique of reforestation using two pine species (*Pinus sylvestris* and *Pinus nigra*) as targets. Previous results showed that shrubs facilitated the early survival of pine seedlings during the first two growing seasons (Castro et al. 2002). The objective of the present work was to analyze the effect of shrubs on the survival and growth of pine seedlings 4 years after planting, considering the effect of both microhabitat and herbivores. This time span may be considered long enough to evaluate establishment success in reforestation and, thus, to draw robust conclusions concerning the suitability of this alternative technique of reforestation. On the basis of a theoretical framework of plant–plant interactions under harsh physical conditions (Callaway 1995), we hypothesized that shrubs provide regeneration niches, thereby increasing reforestation success by ameliorating the microclimate and by protecting seedlings against ungulate herbivores.

## Materials and Methods

### Study Area

The study site was located 1,800–1,850 m above sea level on the northwestern side of Loma de los Panaderos (Trevenque area, Sierra Nevada, southeast Spain, 37°5′ N, 3°28′ W). Plant cover at the site is patchy, with roughly 36% bare soil, 28% cover by the shrubby evergreen *Salvia lavandulifolia* (a scrub reaching some 25 cm in height), and 10% deciduous spiny shrubs, mostly *Prunus ramburii*, *Crataegus granatensis*, and *Berberis hispanica* (which reach around 150 cm in height) (see Castro et al. 2002 for details on habitat structure). The climate is continental Mediterranean, with hot, dry summers and cold winters. Mean annual rainfall is 830 mm, mean temperature of the coldest month (January) is 3.5°C, and mean temperature of the hottest month (August) is 21.6°C (data from 1990 to 2000). Rainfall in 1997, 1998, 1999, and 2000 was 1,283, 632, 755, and 1,045 mm, respectively. In these areas, forests are formed by the Sierra Nevada Scots pine *Pinus sylvestris*

L. var. *nevadensis* Christ and the Salzmannii's black pine *Pinus nigra* Arnold ssp. *salzmannii* (Dunal) Franco.

### Experimental Design

Two-year-old seedlings of *Pinus sylvestris nevadensis* and *Pinus nigra salzmannii* were planted during March 1997 in different microhabitats (treatments) and monitored until the fourth growing season. The microhabitats were the following: (1) Open, seedlings planted in areas of bare soil; (2) Salvia, seedlings planted under the canopy of individuals of *S. lavandulifolia*; (3) Shrub N, seedlings planted just below the northern edge of the canopy of spiny shrubs (*P. ramburii*, *C. granatensis*, or *B. hispanica*); and (4) Shrub S, seedlings planted just below the southern edge of the canopy of spiny shrubs. Planting points were distributed in three plots of circa 6,000 m², some 200 m apart. One half of each plot was protected from ungulates (Spanish ibex *Capra pyrenaica* in the study area) with a fence 1.5 m high. In each plot, we planted 50 seedlings of each pine species per microhabitat inside the fence and the same number outside the fence (1,200 experimental seedlings per pine species in total: 50 sampling points × 3 plots × 2 herbivory levels × 4 microhabitats), sampling points being randomly assigned. Planting holes were dug 40 cm deep with a mechanical auger (30 cm in diameter for Open and 12 cm for the rest of microhabitats). The reforestation method employed in Open treatment was the usual procedure followed in the region. At the end of June 1997, before the onset of summer drought, pines were examined, excluding from the experiment those that had died due to transplant shock (<5%). The resulting final number of monitored seedlings was 1,115 for Scots pine and 1,146 for black pine. See Castro et al. (2002) for additional details on planting procedure and experimental setup. Fenced areas in the experimental design were used to analyze the effect of herbivores and their interaction with the microhabitat on reforestation success, given that ungulate herbivory is a major reason for reforestation failures worldwide (García-Salmerón 1995; Savill et al. 1997; Riley & Jones 2003). However, herbivore damage was low during the study period for several reasons and did not affect survival. Therefore, data from the fenced and unfenced area of each plot were pooled for the analysis of survival.

### Pine Measurements

For each planted pine, we recorded the following: (1) survival, which was sampled at the end of the summer period (October) and at the end of the winter period (May) from 1997 to October 2000; (2) cause of mortality, which was attributed to (i) drought, seedlings that dried out during the summer without any visible damage, (ii) trampling, seedlings that died from ungulate trampling, (iii) herbivory, seedlings that died from ungulate herbivores, (iv) vole tunnels, seedlings with roots disturbed by

vole tunnels, and (v) frost heave, seedlings lifted and loosened from the soil by winter frost; (3) growth, estimated as the length increase of the leader shoot after each growing season; (4) risk of herbivory (only for seedlings outside the fences), percentage of pines that suffered at least one event of ungulate damage over the study period; (5) damage intensity (only for seedlings suffering herbivory), proportion of the shoots consumed by ungulates, averaged over the years of study while seedlings were alive—this allowed the use of seedlings that died before the end of the experiment; and (6) type of damage, distinguishing herbivory in the leader shoot from that in lateral shoots.

### Data Analysis

Seedling survival was analyzed using a failure-time approach, which measures the time to failure (death) of each individual (Fox 1993). We used Cox's proportional hazards semiparametric model that produces estimates of regression models with censored survival data using maximum partial likelihood as the estimation method (Fox 1993; Allison 1995). In addition, cumulative survival after four growing seasons was compared among microhabitats and for any plot independently, with a contingency analysis to explore the final result without the influence of the shape of the survival curve. The growth of the leader shoot was analyzed using a mixed two-way ANOVA, with year as a random factor and microhabitat as a fixed factor (all plots pooled) and all herbivore-damaged pines being previously excluded. Risk of herbivory was analyzed using a contingency analysis, and damage intensity was analyzed using one-way ANOVA after angular transformation of data (all plots pooled because of low herbivore incidence). Analyses were performed using JMP 5.0 software. Results are expressed as mean ± SE throughout the paper.

### Results

The overall mortality rate was 83.2% for *Pinus sylvestris* and 57.5% for *Pinus nigra* after four growing seasons (all microhabitats pooled; see Figs. 1 & 2). Drought was the main cause of mortality (85.6% of dead seedlings), frost heave was the cause for 14.0% of dead seedlings, while only 19 seedlings died because of other causes (vole tunnels, trampling, and ungulate herbivory). The highest mortality occurred during the first summer, 46.8% of the *P. sylvestris* seedlings and 29.3% of the *P. nigra* dying (Figs. 1 & 2). The microhabitat had a significant effect on the two pine species (L-R $\chi^2 \geq 51.82$, $p < 0.0001$), and the pot also had a significant effect for *P. nigra* ($p < 0.0001$). There was a significant ($p < 0.0001$) plot × microhabitat interaction in the two pine species as a result of differences in survival patterns among plots (Figs. 1 & 2), but in any case the overall pattern of survival was clear cut. Salvia showed the highest survival rates after 4 years of study (28.4% for *P. sylvestris* and 64.4% for
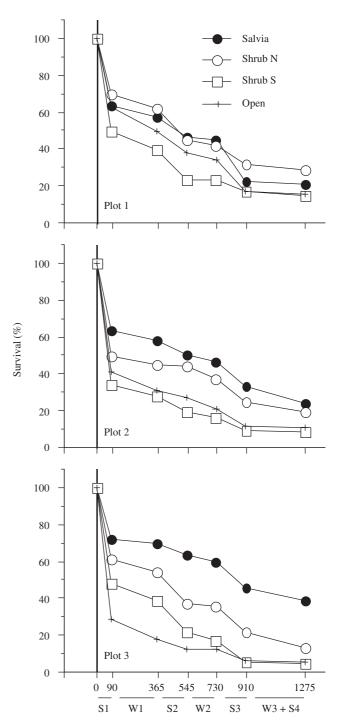


Figure 1. Percentage of *Pinus sylvestris* seedling survival at different sampling periods for each plot and microhabitat. *x*-axis represents days from the start of the experiment (June 1997). S, sampling at the end of the summer period (S1, summer 1997; S2, summer 1998; S3, summer 1999; S4, summer 2000); W, sampling at the end of the winter period (W1, winter 1997–1998; W2, winter 1998–1999). Sampling of winter 1999–2000 (W3) was not done.

*P. nigra*; all plots pooled), Shrub N showed intermediate values (20.3% for *P. sylvestris* and 40.4% for *P. nigra*), whereas Shrub S (9.4% for *P. sylvestris* and 30.2% for
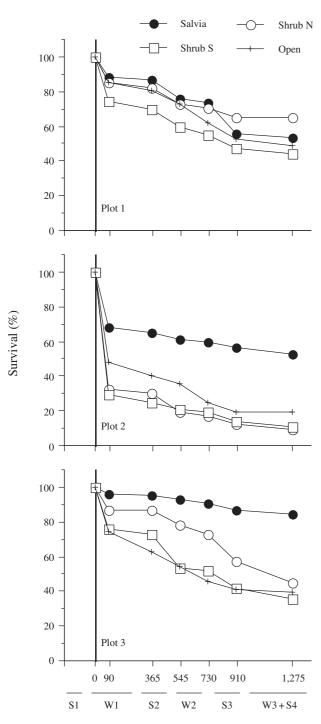
Figure 2. Percentage of *Pinus nigra* seedling survival at different sampling periods for each plot and microhabitat. *x*-axis represents days from the start of the experiment (June 1997). S, sampling at the end of the summer period (S1, summer 1997; S2, summer 1998; S3, summer 1999; S4, summer 2000); W, sampling at the end of the winter period (W1, winter 1997–1998; W2, winter 1998–1999). Sampling of winter 1999–2000 (W3) was not done.

*P. nigra*) and Open (10.7% for *P. sylvestris* and 36.2% for *P. nigra*) registered the lowest values (Figs. 1 & 2). Similarly, differences among microhabitats persisted in all the plots

when considering the cumulative values after 4 study years (*P. sylvestris*: $\chi^2$ from 6.79 to 54.96, $df = 3$, $p = 0.078$; *P. nigra*: $\chi^2$ from 9.68 to 62.70; $df = 3$, $p < 0.05$).

The growth of the leader shoot differed among years. For *P. sylvestris*, the highest growth occurred in 1999, whereas for *P. nigra* there was a steady decrease in the growth of the leader shoot throughout all the years. In any case, there were no differences in growth among microhabitats for each of the pine species in any year. The cumulative growth after four growing seasons (sum of 1997–2000 period) was $18.8 \pm 0.6$ cm for *P. sylvestris* and $26.6 \pm 0.4$ cm for *P. nigra* (all plots pooled; differences between pine species, $p < 0.0001$).

Herbivore damage was low in the two pine species. Risk of herbivory was 8.1% for *P. sylvestris* and 12.7% for *P. nigra* (all microhabitats pooled), with no differences among microhabitats ($p > 0.05$). Damage intensity was $16.8 \pm 4.2$ for *P. sylvestris* (i.e., pines suffering herbivory lost 16.8% of their shoots) and $21.4 \pm 4.0$ for *P. nigra*, with no significant differences among microhabitats for any pine species. When a pine was damaged, the leader shoot was consumed in 89.7% of the cases, and the rest of the damage was in lateral shoots.

## Discussion

The results show that shrubs did not reduce the survival of planted pines but rather clearly enhanced seedling establishment. After 4 years, the survival for *Pinus sylvestris* and *Pinus nigra* under the canopy of *Salvia* was 2.6- and 1.8-fold higher, respectively, than the values reported for the traditional technique in areas of bare soil. This resulted despite planting conditions being a priori more favorable for pines planted in the open areas (larger hole size and additional working around the seedling; Castro et al. 2002). A similar facilitative effect of shrubs on woody plants' seedling survival has been reported for plantings or sowings in Mediterranean-type ecosystems, by, for example, Rousset and Lepart (2000) in southern France and by Callaway and D'Antonio (1991), Callaway (1992), and Dunne and Parker (1999) in California (see also Maestre et al. 2001 for a facilitative effect of tussock grass in semiarid southeast Spain). Furthermore, this positive effect on reforestation has been recently reported for southeast Spain with a meta-analysis considering a wide range of nurse shrubs, target species, and local environmental conditions (Gómez et al. in press). In addition, similar facilitative effects of shrubs on seedling survival in woody plants have been reported for other ecosystems having a dry period, as, for example, Chambers et al. (1999) in semiarid pinyon and juniper woodlands of western North America, Li and Ma (2003) in mixed oak forests of northern China, or Rodríguez-Trejo et al. (2003) in pine forests of the southeastern United States. Therefore, the use of shrubs as nurse plants to boost reforestation success emerges as an alternative technique not only

for Mediterranean-type ecosystems but also for other water-stressed environments.

The first growing season was the most critical period for survival, accounting for more than 50% of the deaths. In addition, the pattern of survival across microhabitats was established mostly during the first growing season, with the rank order of among-microhabitat survival rates remaining similar after 4 years for the two pine species. This supports the contention that the first year is the most critical period for the establishment of planted seedlings (García-Salmerón 1995; Savill et al. 1997; Rey-Benayas 1998), confirming the robustness of conclusions drawn from this 4-year study. On the other hand, shrubs did not reduce the growth of the leader shoot in any of the study years. This supports the effectiveness of this reforestation technique that enhances survival without hampering seedling performance.

The low ungulate damage during the study may have several explanations. First, these pine species have low palatability, and the patch is furthermore dominated by *Salvia lavandulifolia*, which is not attractive to ungulates (Zamora, unpublished data). Second, reforestation took place in a sector of the National Park free of domestic livestock (sheep and goats) and in addition had a low population of the Spanish ibex during the study years. And third, damage intensity on juveniles increases with sapling size (Zamora et al. 2001), meaning that the small size of these saplings might not attract the attention of ungulates. In any case, most of the herbivory was on the leader shoot, which considerably slows down pine growth (Hódar et al. 1998; Zamora et al. 2001). These species, particularly slow growing under the dry-summer Mediterranean conditions of the study area, require around 25 years for the leader shoot to grow beyond the reach of ungulates (circa 150 cm; Zamora et al. 2001). Therefore, even the low herbivore damage registered here may imply that around 60% of pines will undergo herbivory before reaching this height threshold (i.e., average of 10% pines browsed in 4 years × 6). In fact, in sectors of these mountains with higher wild and domestic ungulate pressure, most of the juvenile pines suffer herbivory (Zamora et al. 2001). Under such circumstances, tree seedlings surrounded by shrubs are protected from ungulates in the study area (García et al. 2000; Gómez et al. 2001a, 2003; see also Callaway & D'Antonio 1991, Rousset & Lepart 2000 for other study systems). All this strongly reinforces the contention that nurse shrubs may protect reforested seedlings against ungulates, provided that seedlings are surrounded by shrubs.

The beneficial effect of shrubs on seedling survival is presumably due to the improved water status of the seedlings through reduction of radiation, lowering soil temperature, and conserving soil moisture (Gómez et al. 2001b; Castro et al. 2002). This is further supported by the fact that survival on the south side of shrubs (where microclimatic conditions were close to those in Open microhabitat; Castro et al. 2002) was similar to the survival

in the Open microhabitat, in contrast to the higher values for the north side of shrubs, supporting a direct effect of shading (Valiente-Banuet & Ezcurra 1991; Kitzberger et al. 2000; Rousset & Lepart 2000). Experimental field studies indicate that the shade provided by the canopy of shrubs or artificial meshes is the main factor favoring the survival of planted seedlings in the same study area (Gómez et al. 2001b; see also Callaway 1992, Rey-Benayas 1998, Dunne & Parker 1999, Weltzin & McPherson 1999, Shumway 2000, Rey-Benayas et al. 2002 for a similar canopy effect). Therefore, despite the heliophillous nature of these pine species (Ceballos & Ruiz de la Torre 1971; Nikolov & Helmisaari 1992), the high radiation in Mediterranean environments can impair seedling performance, which may be improved by a moderate light reduction, lowering the risk of photoinhibition and leaf overheating that decreases growth or survival (Valladares 2003). All this suggests that shrubs such as *Salvia*, characterized by a shallow root system and a short, open canopy that moderately reduces radiation, may potentially provide the best functional traits to ensure reforestation success (Gómez et al. in press). The nurse plant will provide protection during the first years after planting, thereafter being outgrown by the sapling (Fig. 3). *Salvia* species as well as other shrubby species with similar morphological characteristics are common in degraded habitats of Mediterranean mountains, and therefore, the use of this reforestation technique could be extensively employed. In any case, we should consider that seedling survival after 4 years was still low in the case of *P. sylvestris* (e.g., roughly 28% in the *Salvia* microhabitat). The technique could therefore be used in combination with additional reforestation techniques for species having low survival rates, in particular with procedures directed to reduce drought stress, such as application of water-retention gels and tree shelters or digging holes of larger size.

The alternative technique of reforestation proposed here emerges from in situ studies of natural regeneration showing that the regeneration niche of woody species in the Mediterranean mountains is positively associated with shrubby species (Rousset & Lepart 1999; Castro et al. 2004; García et al. 2000; Gómez et al. 2001a, 2003a). Under the stressful conditions characterized by summer drought and herbivore pressure, competition is compensated by protection against environmental stress, resulting in net facilitation (Bertness & Callaway 1994; Callaway 1995; Brooker & Callaghan 1998). This facilitative principle derived from ecological studies is therefore a necessary tool for the development of suitable silvicultural strategies for forest regeneration under Mediterranean scenarios. The technique mirrors natural, ecological succession, favoring the transition from pioneer shrublands to mature forests. In addition, a spontaneous, natural shrub cover is already present in most degraded areas where reforestation is required, ensuring the applicability of the technique. All this translates into economic (Castro et al. 2002) and, particularly, ecological benefits, reducing the visual

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Figure 3. A sapling of *Pinus nigra* outgrowing *Salvia lavandulifolia* 4 years after planting.

impact of the reforestation, helping create spatially non-uniform and more diverse stands that are more resistant to pathogens, herbivores, and climatic hazards (Watt 1992; Schönenberger 2001; Hódar et al. 2003). This approach also largely avoids the problems of erosion caused by traditional, more aggressive techniques that eliminate the shrub cover using heavy machinery (García-Salmerón 1995; García-Latorre 1998; Linares et al. 2002; Pardini et al. 2003). These advantages are particularly relevant in protected areas where conservation and minimization of impact are a major priority or in mountain areas with complex orography.

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