Applied Vegetation Science 9: 137-142, 2006

Restoring *Quercus pyrenaica* forests using pioneer shrubs as nurse plants

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Abstract

Question: How to improve reforestation success of *Quercus pyrenaica*.

Location: 1800 m a.s.l., southern Spain.

Methods: One-year-old *Quercus pyrenaica* seedlings were planted using two treatments: (1) bare soil, using a 30-cm diameter augur bit (conventional technique) and (2) under the canopy of a pioneer shrub, *Salvia lavandulifolia*, using a 12-cm diameter augur bit. Survival and growth were monitored for six years. Our hypothesis is that the use of shrubs as nurse plants is an alternative technique of reforestation with higher success than traditional techniques, in which pre-existing vegetation is usually considered a source of competition. The rationale for the study was that for environments with a dry season, pre-existing vegetation buffers summer drought stress, ameliorates the water status of seedlings and thus usually increases seedling recruitment.

Results: *Quercus* survival was 6.3 × higher when planted under individuals of the pioneer shrub as compared to open areas. *Quercus* seedlings under shrubs also had shoots 1.8 × longer, while the number of shoots per plant did not differ among treatments. The first summer was the period with the highest mortality (49.1% of seedlings). Summer drought was the main cause of mortality.

Conclusions: The use of shrubs as nurse plants for *Q. pyrenaica* reforestation is a viable technique to increase establishment success. The technique could be similarly useful in other environments with a dry period and for other *Quercus* species. In addition, this technique offers the advantage of following natural succession, thus minimizing the impact in the community.

Keywords: Facilitation; Mediterranean ecosystem; Reforestation; Seedling establishment; Silviculture.

Introduction

Quercus pyrenaica (Melojo oak) is an abundant deciduous species of the western Mediterranean basin, extending through western France, the Iberian Peninsula and northern Morocco (Franco 1990). *Q. pyrenaica* is a major tree in Mediterranean silviculture (Jiménez et al. 1998; Costa-Tenorio et al. 1998), but these forests are now seriously deteriorated due to historical human action (Calvo et al. 2003; Ibáñez et al. 1997; Blanca et al. 1998; Blanca 2002), and in many cases they represent impoverished patches of former, more extensive forest ecosystems (Gutiérrez de Loma 2001). In addition, after perturbation *Q. pyrenaica* forests may be replaced by sclerophyllous *Quercus* species such as *Q. ilex* and *Q. suber*, which are better adapted to the poorer soils and the drier conditions once the original forest has disappeared (Ibáñez et al. 1997). In fact, intensive human intervention has probably caused the complete replacement of *Q. pyrenaica* forests by other communities in recent times, which is particularly relevant for populations located at the southern boundary of the species distribution area (e.g. southern Spain; Ibáñez et al. 1997; Blanca 2002).

Natural regeneration of *Q. pyrenaica* forest is seriously constrained by several factors, particularly at its southern locations. First, the species reproduces mostly by resprouting and acorn production is low and scattered (Jiménez et al. 1998; Castro et al. unpubl.). Second, acorns, when available, suffer extremely high rates of predation by several vertebrates, reaching losses that effectively prevent regeneration (Mesón & Montoya 1993; Gómez et al. 2003). In any case, acorns are highly predated even after sowing (Gómez et al. 2003), and thus the most common procedure for reforestation is the planting of seedlings grown in nurseries (Mesón & Montoya 1993; Montoya 1997). However, plantations of *Q. pyrenaica* suffer heavy losses, primarily from summer drought, that destroy restoration efforts (e.g. Gómez et al. 2003; Fernández-Abascal et al. 2004). These losses, similarly common for other *Quercus* species as well as other reforested trees in the Mediterranean area (García-Salmerón 1995; Vallejo et al. 2003; Castro et al. 2002, 2004a; Gómez-Aparicio et al. 2004), are caused mostly by heavy mortality during the first growing season (Vallejo et al. 2003; Castro et al. 2002, 2004a; Gómez-Aparicio et al. 2004).

In this work, we experimentally tested the usefulness of an alternative technique of reforestation for *Q. pyrenaica* by considering the use of pioneer shrubs as nurse plants. This technique offers the advantage of a reduced impact on the ecosystem in relation to traditional techniques of reforestation, and has proved very effective in Mediterranean type ecosystems (e.g. Castro et al. 2002, 2004a; Gómez-Aparicio et al. 2004). For this purpose, the survival and growth of *Quercus* seedlings planted in open spaces following a traditional technique were compared with values found for seedlings planted in association with shrubs. The experimental study was monitored for six years, a time span long enough to evaluate establishment success in reforestations and thus to draw robust conclusions about the suitability of the technique. The rationale for the study was that, for dry environments such as the Mediterranean, the presence of nurse shrubs may increase establishment success by reducing summer drought stress (Callaway 1995; Castro et al. 2002, 2004a, b; Gómez-Aparicio et al. 2004), resulting in facilitation. We hypothesize, therefore, that pioneer shrubs will provide a regeneration niche for *Q. pyrenaica* seedling establishment. Three specific questions were posed: 1. What is the effect of shrubs on survival and growth of reforested seedlings? 2. Are the patterns of survival and growth coupled? and 3. What is the most critical period for the establishment of planted seedlings?

Material and Methods

Study site and species

Quercus pyrenaica forests occur mostly between 400 and 1600 m a.s.l., rising above 1800 m in southern locations, and require between 650 and 1200 mm annual precipitation. The species grows predominantly in siliceous soils, but forests are also found on calcareous soils in localities where precipitation is high enough to allow a soil pH to fall close to a neutral value (Franco 1990; Costa-Tenorio et al. 1998).

Q. pyrenaica forms soils of high quality, which, in turn, has implied a drastic reduction of its distribution area in favour of agricultural lands (Costa-Tenorio et al. 1998). The species resprouts abundantly from a dense net of shallow roots, which allow rapid regeneration after perturbations and make it a suitable tree species for coppicing and preventing erosion (Costa-Tenorio et al. 1998; Gutiérrez de Loma 2001).

Experimental reforestation was carried out at 1800-1850 m a.s.l. on Loma de los Panaderos (Trevenque area, Sierra Nevada, SE Spain, 37°5' N, 3°28' W), in the altitudinal belt of *Q. pyrenaica*. The site was a gap of ca. 8 ha occupied by a successional shrubland that had regenerated naturally after a fire in 1983. Today, the area is dominated by *Salvia lavandulifolia* (28.3% cover), an evergreen with a maximum height of 50 cm, intermingled with areas of bare soil or soil with scattered herbaceous vegetation (see Castro et al. 2002 for details on habitat structure). The slope is 5-15°, and the site is oriented towards the northwest. The bedrock is calcareous and the predominant soils are regosols and cambisols (Delgado et al. 1989). The climate is subhumid Mediterranean, with rainfall (860 mm annual mean; 1990-2002) heaviest in autumn and spring, alternating with hot, dry summers and cold winters. The mean minimum temperature of the coldest month (January) is –1.2°C and the maximum mean temperature of the hottest month (July) is 28.5 °C. Rainfall during the study years was 632, 755, 1045, 1054, 1106 and 975 mm in 1998 to 2003, respectively. Livestock has been forbidden in the study site since 1997 (when the area was declared a National Park), which caused low herbivore damage to planted seedlings.

Experimental design

One-year-old *Q. pyrenaica* seedlings were planted in March 1998 in two treatments (microhabitats): (1) Open: seedlings planted in areas of bare soil and (2) *Salvia*: seedlings planted under the canopy of *Salvia lavandulifolia*, which was used as a pioneer shrub. The technique used for planting in the Open microhabitat employed the usual procedure followed in the region, consisting of planting seedlings in open interspaces to avoid proximity to shrub species. The planting holes were dug 40 cm deep with a mechanical augur with a 30-cm diameter bit. This is the augur traditionally used in this type of reforestation in the region, as it provides the greatest hole without hampering its manipulation by workers due to excessive weight. After planting, the soil was manually worked in an area of 0.5 m^2 around the plant to improve soil structure and increase water retention (Mesón & Montoya 1993; García-Salmerón 1995; Savill et al. 1997). For planting in the *Salvia* microhabitat, we used a smaller augur bit (12 cm diameter) to minimize damage to the shrub roots, and no further cultivation was done around the plants. Initial conditions for establishment and growth were thus poorer in the *Salvia* microhabitat due to the lower volume of loose soil and lack of further cultivation around the plants (García-

Salmerón 1995; Savill et al. 1997). In all cases, the soil was returned to the holes and firmed around the root collar of the seedling. The *Salvia* plants chosen as planting sites were between 20 and 35 cm high. No further work was done in the reforestation.

Planting points were distributed ca. 200 m apart in three plots of ca. 6000 m^2. In each plot, we planted 100 seedlings per microhabitat (600 experimental seedlings in total), sampling points being randomly assigned. All seedlings were tagged and mapped. At the end of June 1998, before the onset of summer drought, *Quercus* seedlings were examined, and those that had died due to transplant shock (2.6% of planted seedlings) were excluded from the experiment. Seedlings were purchased from a nursery located at 1600 m a.s.l. at Sierra Nevada (Soportújar, Granada), grown in plastic trays (18 cm deep, 4 cm side) filled with a substrate of native soil mixed with organic material. The mean length of the seedlings at planting was ca. 5 cm. The acorns came from local populations of Sierra Nevada National Park. The experiment was monitored until the sixth growing season.

Seedling measurements

For each experimental seedling we recorded: (1) Survival, which was sampled at the end of the growing season (October) in 1998, 1999, 2000 and 2003; (2) Cause of mortality; (3) Length of the longest shoot, measured three (October 2000) and six (October 2003) years after planting; (4) Number of shoots, which was also sampled three and six years after planting. Survival was corrected through the years for those seedlings apparently dead but resprouting the following year (a common phenomenon in this and other *Quercus* species), so that any resprouting seedling was considered alive. Resprouting of aerial biomass was not further recorded after the fourth growing season (see Fernández-Abascal et al. 2004 for similar results), and thus the final survival data analysed (six years after planting) can be considered free of spurious results due to resprouting.

Data analysis

Seedling survival was analysed with a failure-time approach, which measures the time to failure (death) of each individual (Fox 2001). Plot, treatment and their interaction were used in the model. We used the Cox's Proportional Hazards semiparametric model, which produces estimates of regression models with censored survival data using maximum partial likelihood as the estimation method (Allison 1995; Fox 2001). The growth of the leader shoot was analysed using a mixed two-way ANOVA, with year as a random factor and microhabitat as a fixed factor. Number of shoots was analysed with a non-parametric Mann-Whitney test. Seedlings that suffered herbivory during the study period (1.1% of live seedlings) were excluded from the growth analysis. Because seedling survival was very low in the Open treatment (see below), we pooled the data from all the plots for analysis of growth. For the ANOVA, data were log-transformed (Zar 1996), and we used type III sum of squares. Analyses were performed using JMP 5.0 software (SAS Institute). Means ± SE are shown throughout the paper.

Results

Seedling survival differed among treatments (Table 1; Fig. 1). The survival percentage under *Salvia* was 34.8% after six years (all plots pooled), while in the Open microhabitat only 5.5% of seedlings were still alive. The survival rate differed among plots (Table 1) but in all cases followed the same among-microhabitat pattern, with seedlings planted under *Salvia* showing higher survival in all the plots (no plot × microhabitat interaction; Table 1). The highest mortality occurred during the first summer, when 49.1% of the initially established seedlings died (Fig. 1). Summer drought was the main cause of mortality (99.2%), with very few seedlings dying from other causes (ungulate trampling and vole tunnels).

Shoot length differed between years ($P < 0.0001$) and treatments ($P < 0.0001$), and the differences between microhabitats followed the same pattern during the six years (non-significant year × treatment interaction; two-way ANOVA). Thus, seedlings planted in the *Salvia* microhabitat were longer than seedlings planted in Open either three or six years after planting (Fig. 2). The number of shoots did not differ between treatments, either at three or six years after planting (Mann-Whitney *U*-tests, $P > 0.24$).

Table 1. Summary of the Cox's Proportional Hazards semiparametric model testing the effect of plot and microhabitat on *Quercus pyrenaica* seedling survival for a six-year period. Treatments are Open (areas of bare ground) and *Salvia* (under the canopy of *Salvia lavandulifolia* plants).

Source	*df*	L-R χ^2	*P*-values
Plot	2	13.39	0.0012
Treatment	1	40.34	< 0.0001
P × T	2	2.64	0.2672
Model	5	53.30	< 0.0001

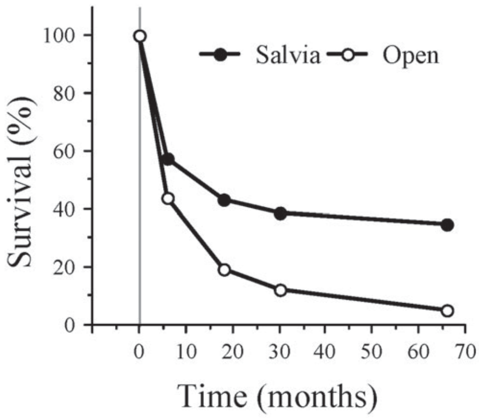

Fig. 1. Percentage of *Quercus pyrenaica* seedling survival at different sampling periods for each microhabitat. X-axis represent months from the start of the experiment.

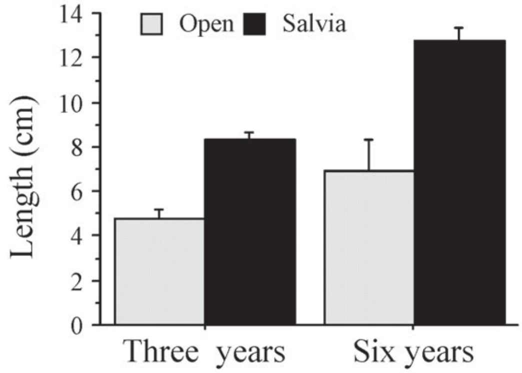

Fig. 2. Length of the leader shoot of *Quercus pyrenaica* seedlings three and six years after planting. Seedlings that suffered herbivory during any year of study were eliminated.

Discussion

Our results show that the survival of *Q. pyrenaica* seedlings was 6.3 × higher under *Salvia lavandulifolia* than in the open microhabitat, despite the fact that the planting conditions were ostensibly more favourable for seedlings planted in the latter microhabitat. This huge difference gains further relevance when considering the percentage of surviving seedlings, as only 5.5% of those planted in the open microhabitat survived after six years in comparison to 34.8% under *Salvia lavandulifolia*. Furthermore, the patterns of growth and survival were coupled, the length of the leader shoot after six years being 1.8 × higher under *Salvia* in comparison to open areas. The growth of the seedlings, although low, was in accordance with those reported for *Q. pyrenaica* even in more favourable areas of its range (Fernández-Abascal et al. 2004).

The first growing season was the most critical period for survival, accounting for approximately half of the deaths. In addition, the survival pattern across treatments was established during the first growing season, with the rank order of among-microhabitat survival rates remaining similar over consecutive years. This supports the contention that the first year is the most critical period for the establishment of reforested tree seedlings (García-Salmerón 1995; Savill et al. 1997; Rey-Benayas 1998), and confirms the robustness of conclusions drawn from this six-year study. Thus, the results show on one hand that the restoration of these *Q. pyrenaica* forests was not possible in open interspaces, and on the other hand that it is necessary to employ a technique that ensures a high survival rate during the first growing season. The extremely low values of survival in the open microhabitat might be influenced by the fact that the year of sowing was relatively dry. This magnifies the relevance of shrubs as nurse plants, allowing survival even in relatively adverse years (e.g. Gómez-Aparicio et al. 2004).

The mechanisms underlying facilitation in Mediterranean type ecosystems are related primarily to a reduction of radiation intensity during summer, which improves the water status of the seedlings through lowering of soil temperature and conserving soil moisture (e.g. Callaway 1995; Dunne & Parker 1999; Castro et al. 2002, 2004b; Gómez-Aparicio et al. 2004). Experimental field studies indicate that the shade provided by the canopy of shrubs or artificial meshes is the main factor favouring the survival of *Quercus* seedlings planted in Mediterranean type ecosystems as well as other water stressed habitats (Callaway 1992; Rey-Benayas 1998; Weltzin & McPherson 1999; Gómez-Aparicio et al. 2005). All this suggests that pioneer shrubs, such as *Salvia*, characterized by a short, open canopy that moderately reduces radiation during the first years after planting, may potentially ensure reforestation success (see also Gómez-Aparicio et al. 2004).

Salvia species, as well as other shrubby species with similar morphological characteristics, are common in degraded habitats of the Mediterranean mountains, and thus the use of this reforestation technique could be extensively employed for *Q. pyrenaica* reforestation. Other possible mechanisms, such as a nutritional improvement of soil under shrubs or a richer soil microbial and fungal communities cannot be ruled out (e.g. Carrillo-García et al. 1999, 2000), strengthening the role of shrubs as regeneration niches for planted seedlings. In fact, soil beneath *Salvia lavandulifolia* has a higher concentration of P and K than in open interspaces (Gómez-Aparicio et al. 2005). In this sense, it is noticeable that mature *Q. pyrenaica* forests grow mostly on siliceous soils. The species is thus traditionally considered to require decarbonated soils, although it is also present in calcareous soils (Costa-Tenorio 1998). Soil properties lose importance in vegetation dynamics with the maturation of the community, given the buffering effect of organic matter on bed rock characteristics. The use of shrubs could thus be particularly critical for the restoration of these forests on calcareous soils. Furthermore, shrubs may protect *Q. pyrenaica* seedlings from browsing and trampling of vertebrate herbivores (Gómez et al. 2003; see also Callaway & D'Antonio 1991; Rousset & Lepart 2000 for other *Quercus* species and García et al. 2000; Castro et al. 2004b; Baraza et al. in press, for other tree species).

The facilitative effect of shrubs upon seedling establishment has been similarly reported for other *Quercus* species around the world in environments characterized by a severe moisture stress during the growing season. For instance, Weltzin & McPherson (1999) report increased establishment of *Q. emoryi* under shrubs in temperate savannas of Arizona and Li & Ma (2003) for *Q. liaotungensis* in mixed *Quercus* forests of northern China. More particularly, in accordance with our results, this facilitative interaction has been consistently reported for Mediterranean type ecosystems, as for instance *Q. douglasi*, *Q. lobata* and *Q. agrifolia* in California (Callaway & D'Antonio 1991; Callaway 1992), *Q. ilex* in southern Spain (Gómez-Aparicio et al. 2004) or *Q. humilis* in grasslands of southern France (Rousset & Lepart 2000). This suggests that the use of shrubs as nurse plants to boost reforestation success of the genus *Quercus* might be applicable to a wide range of sites, at least to those environments characterized by moisture stress during the growing season. In the case of *Q. pyrenaica*, our results clearly show that the use of shrubs as planting sites may help to recover these climax forests with the potential to cover vast areas of the western Mediterranean region.

Acknowledgements. We thank the Consejería de Medio Ambiente, Junta de Andalucía and the Direction of the National Park for permission to work in Sierra Nevada. We are especially grateful to Empresa de Transformación Agraria S.A. (TRAGSA) for carrying out the experimental reforestation. Daniel García, José M. Gómez and Lorena Gómez-Aparico kindly helped with the field work and David Nesbitt looked over the English version of the manuscript. Three anonymous reviewers provided helpful comments that improved the manuscript. This study was supported by a PFPI-MEC grant to J.C. and projects FEDER 1FD97-0743-CO3-02 and HETEROMED (REN2002-04041-CO2-01/GLO).

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Received 4 July 2005;
Accepted 15 November 2005.
Co-ordinating Editor: R. Pakeman.