PART III

Restoration in Practice

In the last chapter of Part II the topic was passive restoration and autoregeneration, eg., through encouraging natural dispersal of acorns by animals and the subsequent colonization of trees. In Part III we address seeding and planting techniques, used for afforestation and the active restoration of cork oak woodlands. Both processes entail several steps. The first steps—germplasm selection and ensuring quality of plant material in the nursery (Chapter 11)—are critical, but later steps, such as site preparation, protection of seeds and seedlings, and other field techniques (Chapter 12), also require careful planning, execution, and monitoring.

In Mediterranean climate regions, drought is the main threat to survival of nursery-raised seedlings. In the critical period just after planting and before seedling roots have colonized the soil outside their containerized soil masses, a few days of drought can be fatal. Therefore, nursery techniques must be tailored to reduce transplant shock and to favor successful seedling acclimation through a combination of biotic and abiotic manipulation techniques. In addition, fine-tuned germplasm selection has still a long way to go to significantly contribute to restoration in practice, especially in the face of climate change and global warming. Similarly, field techniques should be designed to increase water supply to and reduce transpiration from newly planted seedlings.

In Chapters 11 and 12 the reader will find much useful information and discussion of all these issues. However, recall that planning, execution, and monitoring should be flexible and will vary somewhat between sites. No standards can guarantee project success because unpredictable and uncontrollable factors often intervene. Furthermore, as we shall see in Parts IV and V, there are major obstacles to putting these strategies and techniques into action that derive from continental and global drivers of a socioeconomic and cultural nature.

V. Ramon Vallejo

Chapter 11

Germplasm Selection and Nursery Techniques

Maria Helena Almeida, Hachemi Merouani, Filipe Costa e Silva, Jordi Cortina, Roman Trubat, Esteban Chirino, Alberto Vilagrosa, Abdelhamid Khaldi, Boutheina Stiti, Sidi Lotfi El Alami, and V. Ramon Vallejo

In the context of forest transition (Mather 2001) and given its potential economic value, large areas of southwestern Europe have been afforested with cork oak in recent decades (see Chapters 1 and 13, and Color Plate 13a, 13b). However, planting had heterogeneous results, with low survival rates. This suggests the need to improve seed handling and nursery and planting techniques. Afforestation programs must also use suitable genetic material. This may become more critical for success in the context of climate change (see Part V). In the European Union (EU), cork oak reproductive material marketed for forestry purposes is controlled by EU Directive 1999/105/CE. The implementation of this directive at the national level favors the use of high-quality *forest reproductive material (FRM)* that is genetically and phenotypically suited to the site. In this chapter we discuss the selection and handling of cork oak acorns and nursery-grown seedlings. Afforestation is a long-term investment. Therefore, ensuring that high-quality propagation material is available at the appropriate time is crucial to the economic and ecological success of new cork oak woodlands. Also, many existing stands need new plantings to fill gaps and improve age structure.

Germplasm Selection

Afforestation in Mediterranean woodlands, as elsewhere, has often been done the cheapest way, with little concern for the genetic quality of the *planting stock*. But proper choice of acorns or nursery stock can be one of a landowner's most cost-effective decisions because the quality of the acorns or seedlings used largely determines success in the long term.

To choose the most appropriate genetic material for restoration and afforestation, it is important to define *regions of provenance* and carry out provenance field trials because most of the variability for adaptive traits of trees is found at the population level. At present only environmental parameters are used to define regions of provenance. For cork oak, insufficient information is available about the variability and genetic control of adaptive traits. However, in forest areas severely affected by oak decline, randomly distributed trees may present high tolerance, maintaining their robustness and health characteristics (Zobel and Talbert 1984). Therefore, seeds should be collected only in high-quality stands, from healthy trees selected in advance. Because of inbreeding risk, collection from isolated trees should be avoided.

Availability and Quality of Initial Acorn Stock

Cork oak trees often have variable and unpredictable reproduction patterns, with some trees producing seeds only once every two to five years (see Chapter 1). Also, acorns rapidly lose their value through desiccation, infection (e.g., from the fungus *Ciboria batschiana*), and insect attack (e.g., chestnut weevil, *Curculio elephas*) (see Chapter 9). Cold storage can help overcome irregular acorn production and maintain a regular supply of acorns to nurseries, with limited viability loss.

Traditional acorn harvesting usually is spread over a period of several weeks. The risks of seed viability loss, acorn predation by animals, and pregermination increase with time. To avoid these risks and reduce the time and cost of harvest, acorn collection should be planned and performed when massive drop occurs in order to preserve the initial acorn quality. At that time, it is advisable to shake the trees gently by means of a rope wrapped around the fine branches. Acorn color and morphology indicate acorn viability (Color Plate 14): Light brown acorns usually have been on the ground for several days and have become dehydrated, whereas a dark brown color indicates that acorns have been infested by fungi.

Acorn Manipulation, Storage, and Quality Assessment

Cork oak acorns are vulnerable to dehydration, like all acorns. This makes their long-term storage extremely difficult (Stiti 1999; Peñuelas and Ocaña 2000; Montero and Cañellas 2003a). Traditionally, acorns were stored in mesh bags with a simple fungicide application, but this resulted in almost total loss of viability in just three months. A new long-term storage technique (Merouani et al. 2001c) allows storage of cork oak acorns for at least seventeen months without loss of acorn quality (Merouani et al. 2004). The key is efficient organization of all steps in the storage process, starting at the time of seed harvest. The main steps are ensuring initial acorn quality (e.g., maturity, moisture content), choosing the time and type of harvest (controlled or traditional), treating acorns (*thermotherapy* and drying), choosing the type of storage bags, and maintaining storage conditions (temperature and relative humidity) (Merouani et al. 2001a, 2001b).

Successful establishment requires high seedling survival and growth, which are affected by seed quality. Many countries have adopted seed certification as stipulated in Article 14 of the EU Directive 1999/105/CE, which standardizes seed quality in terms of origin, genetic traits, and purity of seed lots. Accurate seed characterization and good seed management decisions require thorough knowledge of the characteristics of the product. This evaluation procedure entails acorn sampling in order to guarantee reliable data. Seed lot samples must be truly representative of the whole lot, and competent analysts must carry out the testing.

Plant Production and Nursery Practices

Large-scale cork oak planting may have occurred as early as the end of the nineteenth century (Natividade 1950). For example, at Rio Frio, Portugal, 3,600 hectares was planted using cork oak seedlings in wooden boxes, and yet as recently as thirty years ago, seedling production was still unusual in forest nurseries. Instead, acorns were seeded directly despite incompatibility with agroforestry practices and the unpredictable supply of acorns. Over the last fifteen years, however, oak seedlings in containers have become more common, and greater attention has been paid to improving quality and developing seedling certification procedures.

Planning of seedling production is an essential step to achieve quality, and field performance is a critical part of planning (Landis 1993). Nursery managers must be informed ahead of time about the area to be planted, the location, and site characteristics. Furthermore, nursery cultivation regimes can strongly affect the functional characteristics of seedlings (i.e., their quality) and consequently their field performance (Villar-Salvador et al. 2004). This depends on the proper use and timing of cultural practices, including sowing date, type of container, growing substrate, and watering and fertilization regimes (Vilagrosa et al. 1997; Cortina et al. 2004). Both the storage of acorns in inappropriate conditions and the inadequate manipulation of

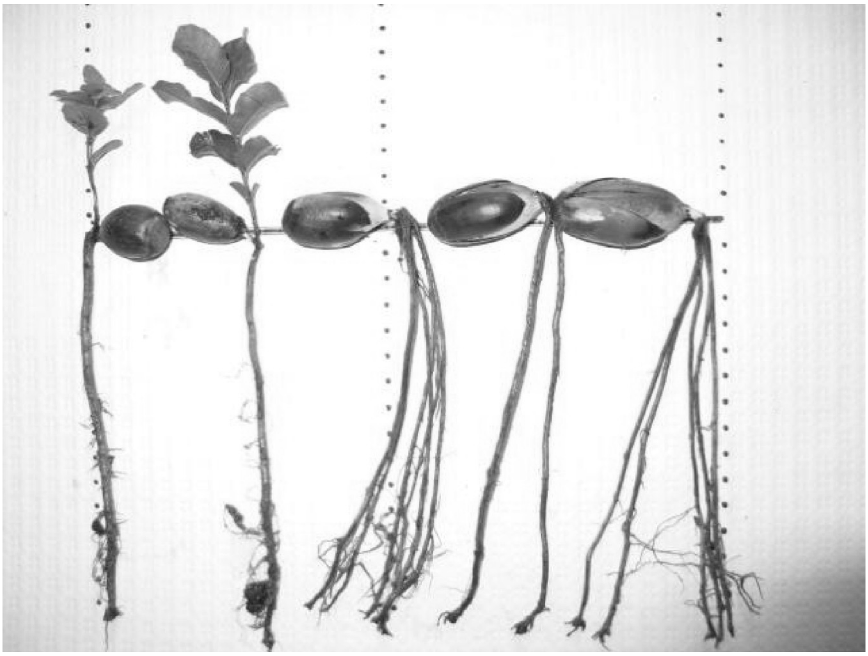

FIGURE 11.1. Cork oak seedling development with acorns showing typical roots *(left)* and fasciculated root systems *(right)*.

pregerminated acorns result in high failures of seedling emergence, such as 30 to 40 percent losses (Peñuelas and Ocaña 2000). This is often caused by damage of pregerminated acorns at the radicle–shoot junction during sowing. Figure 11.1 shows cork oak seedlings with a typical taproot and seedlings with a fasciculated root system, with two or more main roots rather than a single one, as a result of radicle–shoot breakage. Sowing controlled, pregerminated seed typically yields more than 85 percent seedling emergence and avoids the problem of shoot weakness.

Finally, seedlings with high aboveground biomass and low root:shoot ratios suffer more intense transplant shock, and consequently higher mortality and lower growth in the field (Costa e Silva et al. 2001).

Seedling Age

In traditional tree nurseries, the planning of seedling production is limited because acorns are available for only a short time (typically November and December). However, improved storage techniques allow year-round acorn availability. Another advantage of storage is that it accelerates acorn germination and seedling emergence rates, increases the *relative growth rate (RGR)*, and increases seedling uniformity (Merouani et al. 2001b, 2005).

Sowing acorns immediately after harvest and planting the next autumn leads inevitably to a longer period in the nursery (and additional seedling costs), which may negatively affect their root–shoot equilibrium. Furthermore, fresh acorns show wider variation in the time of germination than cold-stored acorns (Merouani et al. 2001b). This can create serious nursery crop problems related to variable seedling sizes because small initial differences in size can lead to large size differences at the time of shipping and cause difficulties with oak seedling certification (EU Directive 1999/105/CE in Annex VII Part E).

Containers and Substrates

New types of containers for growing seedlings are continuously being developed to reduce handling costs and improve seedling quality. Indeed, container design largely determines the morphological and physiological characteristics of tree seedlings (Aphalo and Rikala 2003; Villar-Salvador et al. 2004; Pemán et al. 2006). Container volume and height affect nutrient availability and space for root development. Container diameter has a direct influence on distance between seedlings, and therefore competition for light, and on the relationship between shoot and root growth, and therefore stem diameter. Although there is no standard type of container in use, 300 cubic centimeters is the most commonly used volume in the Iberian Peninsula (Costa e Silva et al. 2001; Montero and Cañellas 2003a; see Color Plate 13c). In southeastern France, 600–cubic centimeter containers with anticoiling interior ribs are preferred and have proven useful (P. Brahic, personal communication, 2006). Containers coated with copper carbonate may also help produce smaller seedlings and avoid spiraling roots (Pardos et al. 2001). Larger containers induce higher seedling growth (Domínguez et al. 1997; Suárez et al. 1997). However, seedlings cultivated in deep containers acquire several morphofunctional advantages, improving seedling water status under drought conditions (Chirino et al. 2008), although the differences in seedling size often disappear over time in the field because growth is no longer constrained by the amount of space and the substrate provided by the container (Costa e Silva et al. 2001).

The growing substrate provides physical support for the seedling, and it should provide water, nutrients, and air. There are many different types of substrates on the market that have been used successfully for cork oak seedling production (Figure 11.2), including granulated cork, a byproduct of

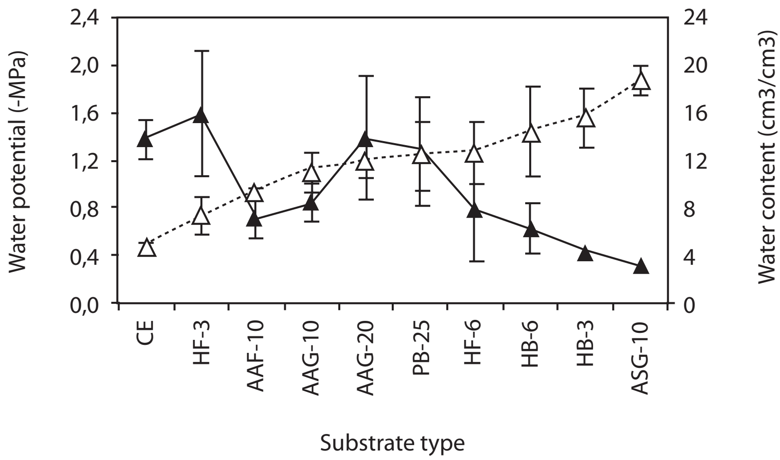

Figure 11.2. Changes in water status and substrate moisture content in cork oak seedlings planted in different growing mixes and subjected to a 7-day drying period. Bars correspond to means ± *SE*. AAF, fine atapulgite (10% and 20% v/v application rates); AAG, coarse atapulgite (10% v/v application rate); ASG, coarse sepiolite (10% v/v application rate); CS, control mix (peat and coconut peat, 50:50 v/v); HB, medium hydrogel Bures (0.7% and 1.5% w/w application rates); HS, medium hydrogel Stockosorb (0.7% and 1.5% w/w application rates); PB, pine bark (25% v/v application rate). All amendments were added to the CE mix. *White triangles*, growing mix water content; *black triangles*, plant water potential. (Data from Chirino and Vilagrosa 2006)

the cork industry itself (Costa e Silva et al. 2001; Merouani et al. 2005). Growing substrates may have a significant effect on cork oak morphofunctional traits and field performance that may last for more than three years. In general, the inclusion of substrates that improve water retention and availability, such as peat, promotes higher field performance (Landis et al. 1990; Costa e Silva et al. 2001). However, the use of pine bark and soil organic layers in substrate leads to poor performance, which can be explained by unfavorable water retention properties and difficulties in forming a consistent firm root plug. Field experiments have shown that optimized growing media may increase seedling survival by as much as 30 percent compared with traditional substrates (Figure 11.2).

Adding Fertilizers

Fertilization can accelerate shoot and root growth of seedlings, modify tissue nutrient contents and the amount of available reserves, improve posttransplant rooting and growth capacity, and increase resistance to water stress

(Grossnickle 2000). Several studies have reported a positive relationship between seedling size and nutrient status in planting and field performance (Cortina et al. 1997; Puértolas et al. 2003). We found that heavily fertilized seedlings may double in size, as compared to seedlings receiving no N or P, after ten months in the nursery, whereas the relative amount of biomass allocated to roots did not change. However, cork oaks seedling quality may decline at high fertilization rates. When cork oak tree seedlings were planted in the field, differences in biomass accumulation decreased because part of the aboveground biomass dried in the summer drought. More importantly, differences in size and nutrient content did not generate differences in field survival (Costa e Silva et al. 2001). To date, fertilization in the nursery is not a common practice in cork oak seedling production because growth is initially based on seed reserves, and increased growth can lead to unbalanced plants in the restricted space of small containers over a long nursery growing period. This might be overcome by shortening the production period in the nursery.

Preconditioning Treatments

Seedling hardening or drought preconditioning (i.e., exposing seedlings to periodic stress) has been recommended for acclimatizing nursery seedlings to harsh field conditions. Nevertheless, a recent study with cork oak seedlings found that drought preconditioning had little effect on seedling survival in the field, despite substantial effects on seedling morphology (Chirino et al. 2004).

Nutritional hardening (i.e., reduction in nutrient supply, particularly nitrogen, during the late stages of nursery growth in order to acclimatize seedlings to harsh field conditions) has received less attention than drought preconditioning. Yet seedlings subjected to low N availability may be better adapted to drought because of reduced leaf size and increased allocation of biomass and nutrients below ground (Chapin 1991; Liu and Dickman 1993). Nitrogen availability is often low in degraded Mediterranean soils, and that may limit seedling establishment (Bottner et al. 1995; Martinez-Mena et al. 2002; Valdecantos et al. 2006). Therefore, the depletion of seedling N reserves resulting from a reduction in N application rate could compromise seedling performance in the field. However, a reduction in N supply could favor an accumulation of phosphorus and other macronutrients and improve seedling capacity to withstand low P availability, as is common in many dryland soils (Valdecantos 2003).

Studies of the response of Mediterranean woody species, including cork oak, to N deprivation during their last weeks in the nursery show that foliar N concentration and seedling size are significantly reduced in most species (Trubat et al. 2008). In contrast, the effect of N hardening on seedling survival showed no common pattern. Nitrogen deprivation had a positive effect on survival of seedlings planted under semiarid conditions (Trubat et al. 2008), whereas the effect was not statistically significant in cork oak seedlings planted under dry to subhumid conditions.

Seedling Quality Assessment

Seedling quality, defined as fitness for purpose, is a concept that includes the degree to which trees achieve end-of-rotation goals at a minimum cost (Willén and Sutton 1980). Forest nurseries seek to provide seedling stock for reforestation that can survive prolonged environmental stresses and produce vigorous growth after outplanting. Unfortunately, few studies have been conducted on growth of cork oak seedlings in the nursery in relation to field performance (survival and growth), and no reliable morphological or physiological predictor of plant quality and vitality has yet been identified.

There is an ongoing debate over the optimum seedling size at planting. In the context of the restoration of Mediterranean ecosystems, seedlings should be able to withstand unfavorable growing conditions (transplant shock, summer stress, drought) and still take advantage of favorable weather to grow. Plant investment in root system development is an advantage for field survival because it facilitates water uptake. Conversely, having too many leaves in comparison to roots increases water loss by transpiration without prompt access to water. Nevertheless, high leaf area favors seedling survival as long as it is accompanied by a well-developed root system. In fact, in Mediterranean ecosystems, although soil water may be exhausted from the topsoil during the dry season, enough water is usually available for woody plants in the underlying soil or subsoil, except in extremely dry sites or after severe droughts (see Chapter 6). If roots fail to reach that water because of seedling defects or compact soil layers near the surface, survival is jeopardized. Plant quality indexes, such as relative growth rate (Hunt et al. 2002) and *root growth potential (RGP)*, integrate information about growth processes and are the most reliable parameters available to assess plant quality.

Performance in the field is the ultimate measure of the quality of seedlings used for restoration. Still, the costs of replanting after losses caused by poor seedling quality are so high that often only a small number of seedlings with insufficient growth potential must be identified before plantation to overcome this problem. However, all efforts to produce high-quality seedlings in the nursery will be futile if seedlings are not cared for after they leave the nursery.

Vegetative Propagation

The use of vegetative propagation for cork oak afforestation is not common. This technique is more expensive per plant than seminal production. Furthermore, plant material is not widely available because no comprehensive breeding program has been implemented, and the genetic control of cork is unknown. Bearing in mind that cork oak is a long-lived species and considering climate change scenarios for southwestern Europe and the need to maintain genetic diversity, we believe that the establishment of clonal forests should be avoided. However, studies on vegetative propagation using stem *cuttings* and micropropagation techniques will provide valuable information on cork oak adaptability to changing environmental conditions. If "elite" trees (e.g., tolerant to certain diseases) are identified, these techniques can become appropriate.

Conclusions

Using suitable genetic material by selecting adequate provenances and improving the morphological and physiological quality of reproductive material are necessary steps toward ensuring the success of cork oak plantings, as is the use of appropriate forest reproductive material to offset unfavorable growing conditions. Several innovative nursery treatments lead to improvements in seedling stock quality and performance under field conditions. A key factor is careful planning that includes attention to the entire process, starting from seed collection. The development of long-term acorn storage techniques will facilitate planning and enhance seed and seedling quality. However, there is still a clear need to optimize seedling production in order to reconcile the qualitative and economic aspects of the cork oak restoration and afforestation efforts. Chapter 12 will deal with field techniques used to improve cork oak establishment.

Acknowledgments

The authors warmly thank Isabel Serrasolses for providing helpful information. Financial support was provided by the Portuguese project POCTI/AGG/41359/2001 and Spanish project SUROS (REN2002-11326-E/GLO) and APLITEC (GV 05/208). The Mediterranean Centre for Environmental Studies (Fundación CEAM) is funded by Generalitat Valenciana and Bancaja.

SITE PROFILE 11.1

Aspres and Albères, France

This site is in the eastern part of the Pyrenees, on the Mediterranean coast of southern France, near the Spanish border (French Catalonia). It consists of several patches in which mature mixed forest is regenerating spontaneously.

Geographic and biophysical description	The main location is in the Aspres and Albères formations (15,000 ha), with additional patches in the Têt valley of Roussillon and the Tech Valley (Vallespir). Mean annual precipitation ranges from 500 to 1,000 mm per year. The prevailing winds are the dry and cold Tramontane from the northwest and the humid Maritime from the east. Topography is rugged, and altitude ranges from 0 to 700 m. Bedrock consists of schists, detrital materials of Pliocene (Aspres), and schists, gneiss, and granite (Albères). Accompanying vegetation includes holm oak (*Quercus ilex*), downy oak (*Q. pubescens*), strawberry tree (*Arbutus unedo*), tree heath (*Erica arborea*), rockrose (*Cistus monspeliensis*, *C. salvifolius*), stone pine (*Pinus pinea*), European chestnut (*Castanea sativa*), common smilax (*Smilax aspera*), wild madder (*Rubia peregrina*), broom (*Ruscus aculeatus*, *Calycotome spinosa*), strawflower (*Helichrysum stoechas*), lavender (*Lavandula stoechas*), and false olive (*Phillyrea angustifolia*).
Physiognomic description of the cork oak woodlands and their landscapes, including woodland dynamics	Many pure cork oak forests occur, as well as mixed stands with other oak species. In the absence of active forestry practices, cork oak can be crowded out by other oaks in marginal stands, such as those of the Aspres region. Colonization of abandoned fields or former vines by oaks is also occurring.
History of land uses, land tenure (and socioeconomic drivers), and current land uses, economic activities, and context	Stands were intensively managed in small to medium-sized land holdings from the 18th century until World War II for cork extraction, animal husbandry, and charcoal production. A new wave of tree planting took place at the end of the nineteenth century, on abandoned vineyards devastated by phylloxera root rot. Cork production was abandoned in the 1950s because of competition from Portugal and Spain. Cork extraction was revived in the 1990s, thanks to the rise of cork prices and subsidized projects designed to avoid or reduce the risk of wildfires.
Disturbance regime (fires, pests, overgrazing)	The risk of forest fires has increased since the 1970s, menacing tourist areas and shrublands. This is made worse by low livestock stocking rates, except in strategic zones for forest fire protection, where subsidized grazing is maintained.

	Some cork oak decline has been observed, caused by combined drought and pathogens (e.g., *Platypus cylindrus*, *Diplodia corticola*, *Biscogniauxia mediterranea*). Populations of wild boar (*Sus scrofa*) cause much depredation.
Constraints and conflicts, protective measures, restoration actions, land use regulations, and relevant policies	Recreational hunting, hiking, and gathering of wild berries and mushrooms are increasingly important activities. Forest management, carried out primarily by professional organizations, is not highly intensive. Cork extraction is carried out every 12–15 years, but some problems arise due to bad practices (e.g., tree injuries, cork theft). Some trees have been planted on abandoned old fields and formerly cultivated pastures. Cork oak forests are taken into consideration in local territorial policies.
Current trends and prospects for the future	Cork oak can spread on abandoned lands at low altitudes (<300 m) but tends to suffer in competition with other oaks at higher altitudes (>300 m) or on the plains. Strong pressure exists for housing in areas of contact between urban and forest zones (e.g., the Albères). Cork production is maintained by the presence of local industries in Spanish and French Catalonia.
Source	Renaud Piazzetta, IML (Institut Méditerrannéen du Liège), Vivès, France.

Site Profile Figure 11.1. Argelès-sur-Mer cork oak woodland.