Assessing Morphological and Physiological Plant Quality for Mediterranean Woodland Restoration Projects

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Introduction

Planting seedlings grown in a nursery is often the main way for introducing or re-introducing plant species in woodland restoration projects. If species have been selected properly, the main factors affecting plantation success are environmental conditions, soil preparation and the quality of seedlings (South 2000).

Several reviews on plant quality have been published (see Ritchie 1984, Duryea 1985, Mattsson 1997, Wilson and Jacobs 2006), but most of the experience on this topic has been acquired from humid-temperate and boreal trees. Little information exists on plant quality of species from other biomes. Application of the experience gained in humid-temperate and boreal ecosystems to woodland species from other floras that have different phylogenetic background and environmental constraints than that of humid-temperate and boreal ecosystems must be done carefully.

During the last fifteen years there has been an increase in the study of plant quality, cultivation and plantation techniques of Mediterranean woody species. Our aim in this chapter is to review the procedures and importance for assessing forest plant quality, putting emphasis on recent experience gained with Mediterranean species and highlighting the differences observed *vis à vis* with humid-temperate and boreal forest species. As container stock is the predominant cultivation system of Mediterranean species in the European Union most of the information and examples given here will refer to this type of stock.

Concept of plant quality and importance of its assessment

Plant quality can be defined as the capacity of seedlings to survive and grow after transplanting in a specific environment (Ritchie 1984, Wilson and Jacobs 2005). The survival and growth capacity of a seedling depends on its carbon, water and nutrient economy, which are ultimately determined by the structure and physiological attributes of the plant (Burdett 1990). Plant functional attributes are genetically determined (see Chapter

6, this volume), but they are also plastic phenotypically and can vary depending on resource availability and the environment under which plants grow. The main aim of plant quality research is to determine which structural and physiological properties must seedlings attain for surviving and growing fast in a specific planting environment and how can these functional properties be achieved during nursery cultivation.

The functional attributes that promote seedling success in harsh sites are different than those in mesic sites (van den Driessche 1992). Thus, ideally, plant quality should be adjusted to the characteristics of planting site (Rose et al. 1990). Plant quality may vary over time. For instance, frost hardiness in most temperate species increases from the fall through the winter and then it decreases again through spring in response to changes in photoperiod and temperature (Grossnickle 1992). Thus, assessment of plant quality prior to planting must be done as closer as possible to planting date. Plant quality may also change with plant age. Nicolás Peragón (2004) observed that two-year old *Quercus faginea* seedlings had lower root growth capacity, survival and growth than one-year old plants.

The effect of planting poor quality stock can last for many years or it can be apparent only many years late after planting, as it occurs with root deformations that can reduce tree stability over the long-term (Lindström and Rune 1999).

Plant quality can be assessed by measuring several morphological and physiological attributes (material attributes), or by examining plant performance after subjecting them to specific environmental conditions (performance attributes). The final goal of testing plant quality prior to planting is to prevent plant lots with low survival and growth potential to be planted and to predict the *potential* out-planting performance of planted stock. Plant quality assessment cannot tell us the actual performances of seedlings because out-planting performance also depends on other factors some of which vary stochastically. Assessment of plant quality and use of high-quality plants are important because attainment of restoration and ecological objectives are secured or achieved faster and spread of diseases in natural populations is prevented. Similarly, the final economic cost of restoration projects may be increased if seedlings have to be replanted or if expensive post-planting cares, like irrigation, have to be used to warrant seedling establishment. Nurserymen should produce high-quality seedlings because it warrants consumer confidence and discards that planting failures are due to poor quality stock.

Assessment of plant physiological and performance attributes strongly increases the cost of a plant quality-testing program but it has saved a lot of money in some regions of North America. In British Columbia (Canada), plant quality assessment (physiological quality tests included) amounted 0.4% of the planting program cost (Dunsworth 1997). Assessment of physiological attributes in a plant quality-testing program is worthwhile when planting failures are frequent and planting and seedling cultivation costs are high. It might also be desirable when new species are used in reforestation programs whose biology, cultivation, and transplanting performance are largely unknown.

Material attributes. The morphological component of plant quality

The morphological quality of plants comprises a set of attributes that measure the structure, colour and appearance of the plant. Morphological attributes are the basis of the European Union legislation that regulates plant quality, although specific traits have only been defined for Mediterranean species (Directive 1999/105/CE). Morphological attributes can be either qualitative or quantitative, and most of them can be assessed with simple measurements.

Qualitative morphological attributes

In Table 1 are given the main qualitative morphological attributes used in plant quality assessment together with their rationale, and some cautions to be observed when these attributes are applied to Mediterranean species in a restoration context. The advantage of qualitative morphological attributes is that they are easy to assess. Their disadvantage is that they are to some extent subjective and have limited out-planting predictive capacity. Most qualitative morphological traits have been established from the cultivation and plantation experience accumulated in boreal and humid-temperate tree species. These species have been mainly planted for timber production so possession of specific traits important for timber quality, as single and straight stems, have considered desirable traits in seedlings. Application of certain classical qualitative attributes to shrubs and trees from other biomes or to stock used for restoration purposes might not be straightforward and some adjustment may be needed (Peñuelas and Ocaña 2000). For instance, multi-stem plants tend to be rejected because they produce low timber quality trees but in *Retama sphaerocarpa, a* Mediterranean leafless leguminous species, high-quality plants always produce multiple stems. Similarly, the presence of terminal buds is considered a sign of dormant and cold hardened state. However, in some species cold hardening is not associated with the presence of an apical bud (Colombo et al. 2001). Furthermore, many Mediterranean species have indeterminate growth and do not produce typical resting buds or if they form them they appear in saplings but not in seedlings.

Quantitative morphological attributes

Quantitative morphological attributes are measurements of the shoot and root size. Quantitative traits are used in both scientific studies on plant quality and in operational plant quality assessment. The root collar diameter (RCD), shoot height (from RCD to stem apex), shoot mass, and root mass are the most frequently measured attributes. The number of first order of laterals roots and root volume has also been used in bareroot stock quality assessment. From these measures several indexes have been developed. The most common are the shoot height/Root collar diameter ratio (i.e., shoot slenderness) and the shoot mass/root mass ratio (S/R) that is a proxy to the potential transpiration-water uptake balance of the plant (Ritchie 1984, Thompson 1985, Mexal and Landis 1990, Villar-Salvador 2003). Due to their measurement simplicity, shoot height and root collar diameter are the most

TABLE 1. Main qualitative morphological attributes used in plant quality assessment and their rationale. Some attributes are regulated by European legislation and the lots of plants with more than 5% of individuals with these traits must be rejected.

Attribute	Rationale	Comments on the application to Mediterranean species
Regulated by European Union legislation		
Injuries, except those caused during lifting or by pruning.	This attribute is especially important for bare-root stock. Injured plants have low vigour and poor establishment.	Injuries caused at lifting are rare in container plants but rough handling may damage plant during their transport. Pruning in Mediterranean container nurseries is not a common practice.
Lack of terminal buds	In many boreal and humid-temperate species, dormant and cold hardened seedlings form a terminal bud. Healthy apical buds produce well-developed shoots in spring.	Many Mediterranean species do not form a typical winter bud (a meristem protected by scales). This is the case of some junipers, *Arbutus unedo*, *Pistacia lentiscus*, *Viburnum tinus*, which have naked buds or meristems protected by leaves. Mediterranean pines do not form winter buds until they are saplings.
Multiple stems and main stem bifurcated	Multiple stems may indicate that two or more plants are growing in the same container cell or that the apical meristem bifurcated early, or resprouting has occurred. For timber production, it reduces growth and timber quality, and increases silviculture costs.	Many Mediterranean trees and shrubs have apical dominance as seedlings. However, some species, e.g. some evergreen oaks, can resprout if they are retained two or three years in the nursery. Whether this type of plant has lower out-planting performance is unknown. Some shrubs and many chamaephytes commonly produce multistem individuals.
Deformed root system	Spiralized roots or up growing roots in container plants may reduce new root egress and this may impair early establishment and future tree stability	
Wilted or chlorotic foliage or presence of rottenness or of any disease	Wilted or chlorotic foliage may indicate that the plant is diseased, has experienced severe drought or heat or has a deficient nutrient concentration. Planting infected plants can spread diseases or pests	
Unbalanced shoot root ratio (S/R)	Excessive S/R can cause water stress if new roots are not produced or soil is dry.	In dry areas low S/R is considered a desired trait. However, very low S/R may increase plant maintenance costs and therefore diminish growth
Other important qualitative traits not regulated by the European Union legislation		
Excessive stem curvature	Important for timber plantations because reduces the amount of profitable timber and its quality	Should be avoided if reduces growth and competitive capacity of seedlings
Actively growing shoots and unhardened plants	Plants with growing shoots are not hardened and consequently are less stress-resistant	Important for species planted in cold winter areas.
Lack of branches	The lack of lateral branches in some species may indicate high cultivation density or heavy shading during cultivation	Many species do not produce branches during the first year in the nursery
Poor developed plugs	For container stock poor developed plugs is a symptom of poor root development. These plants experience greater stress during their manipulation and plantation.	

commonly morphological quantitative attributes used for operational quality assessment. Many countries have regulated the shoot height and RCD standards for acceptable seedlings in many planted tree species. For instance, one-year old *Quercus ilex* (Holm oak) seedlings of acceptable quality in any of the Mediterranean countries of the European Union must have a shoot height ranging within 8 to 30 cm and a minimum RCD of 2mm.

Studies done with boreal and humid-temperate species show that plants size has a quite good predictive capacity of out-planting performance, which it frequently increases with shoot and root size, especially in mesic planting sites. Some experiments have shown that seedlings larger than conventional standards can be a promising alternative to herbicides because they compete better with weeds than the smaller conventional stock (Lamhamedi, *et al.* 1998). Relationships of survival and growth with S/R are less clear and often contradictory (Lopushinsky and Beebe 1976, Thompson 1985, Tuttle et al. 1988, Mexal and Landis 1990, van den Driessche 1992, Bayley and Kietzka 1997, South 2000, South et al. 2005). Root collar diameter and plant mass tend to predict better out-planting performance than shoot height and the relationship tends to be stronger with field growth than with survival. However, for plants of the same age there is a size and S/R top limit from which survival and growth plateau and decline (McDonald et al. 1984, Thompson 1985, South et al. 2005).

In Mediterranean environments, water stress is the main limiting factor for plant life and restoration success. This has conditioned Spanish foresters, which have traditionally preferred plants with small shoots and low S/R because they are considered to perform better in dry conditions than large plants and higher S/R (Royo et al. 1997) as they consume less water than plants with the opposite traits (Leiva and Fernández-Alés 1998). This sort of plant was produced with low amounts of fertilizer and frequently by restricting irrigation (Luis et al. 2004). Some evidences support that small plants with low S/R have greater survival than large plants with great S/R in Mediterranean dry areas. For instance, Trubat et al. (2003) observed that small seedlings had greater survival than large plants in *Pistacia lentiscus*. On the contrary, other authors have observed that large plants have higher survival and growth than small seedlings (Fig. 1) (Oliet et al. 1997, Luis et al. 2003, Puértolas et al.2003a, Villar-Salvador et al. 2004, Oliet et al. 2005, Tsakaldimi et al. 2005). A recent revision of 33 experiments on plant quality published by Spanish authors on Mediterranean species does not support the contention that the small seedlings with lower S/R have greater survival and growth than large seedlings and with greater S/R (Navarro et al. 2006). In most cases no relationship was observed between survival or growth and plant size or S/R. When survival was related to morphology, in most cases relationships with shoot size were positive while no trend could be concluded with S/R. Similarly, the results were independent of the type (woodland vs. abandoned cropland) and rainfall regime of the planting site. It was concluded that the present shoot size standards regulated by legislation in several Mediterranean woody trees should be higher (Table 2).

Material attributes. The physiological component of plant quality

Morphological attributes have limited predictive capacity of transplanting performance because they do not tell about the physiological status of the plant, which is also important for plant establishment. For instance, morphology will not indicate if non-structural carbohydrate reserves of a plant are low or its fine roots are damaged. Therefore, physiological quality attributes must be considered as a complement to morphological attributes rather than an alternative. Many physiological attributes have been utilised to assess plant quality (see Mattson 1997). We have focused on those most extensively studied and those that are most promising for plant quality assessment in Mediterranean species.

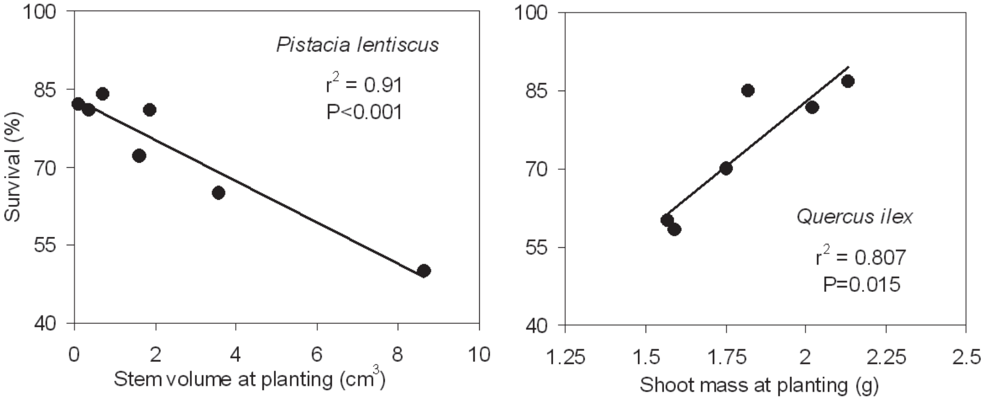

FIGURE 1. Relationship between survival and shoot size at time of planting in *Pistacia lentiscus* (left) and *Quercus ilex* (right). Figure of *P. lentiscus* was elaborated from data published in Trubat *et al.* (2003). Figure of *Q. ilex* is based on unpublished data of P. Villar-Salvador from an experiment with different fertilization and container treatments.

TABLE 2. Proposed standards of shoot height, diameter, slenderness and shoot /root ratios (S/R) for one-year old container seedlings in four common Mediterranean woody species. Values without brackets are proposed ranges and values in brackets are the standards recognized by European legislation. No legal standards exist for *Olea europaea*. (Data adopted from Navarro et al. 2006).

	Shoot height (cm)	Root collar diameter (mm)	Height / diameter (cm mm^{-1})	S/R (g g^{-1})
Pinus halepensis	15 - 30 *(10 - 25)*	3 - 4 *(>2)*	5-7	1.5 – 2.0
Pinus pinea	20-30 *(10 - 30)*	3.5 – 4.5 *(>3)*	5-7	2.0 – 2.5
Quercus ilex	20 - 30 *(8 - 30)*	4-5 *(>2)*	4-7	0.6 – 1
Olea europaea var. *sylvestris*	30 - 50	4-5	7-12	2-4

Mineral nutrient and non-structural carbohydrate concentration

Greater fertilization in the nursery enhances seedling growth and nutrient concentration, which frequently increases field performance (van den Driessche 1992, Villar-Salvador et al. 2004, Oliet et al. 2005). Nitrogen, phosphorus and potassium are the nutrients that most affect plant quality. Nitrogen is the most abundant macronutrient in the plant and it is strongly correlated to photosynthesis rate and growth. Plants remobilize N from old tissues to support new growth after transplanting. Therefore, plants with high N concentration compete better against weeds and have greater growth in oligotrophic soils (Salifu and Timmer 2003). Several studies have shown that post-planting survival and growth in Mediterranean species increases with tissue N concentration (Fig. 2a) (Oliet et al. 1997, Planelles 2004, Villar-Salvador et al. 2004). However, since morphology and tissue N are modified together by N fertilization in the nursery, it is difficult to disentangle the effect of plant size and N concentration on post-planting survival and growth. Plant N content (i.e., the product of dry weight nitrogen concentration by seedling dry weight) was better predictor of post-planting growth in *Pinus halepensis* than seedling dry weight alone (Fig. 2b and 2c). This illustrates that although seedling size determines post-planting growth, tissue N concentration also plays an important role.

Phosphorus forms part of ATP, certain enzymes, and membranes and is involved in the photosynthesis and respiration of the plant. Root growth is especially sensitive to P deficiencies. Effect of P deficiency on plant growth is less obvious than N deficiency. There are few studies linking tissue P to field performance but in semiarid leguminous species, transplanting survival increased with tissue P (Planelles 2004, Oliet et al. 2005).

Potassium is after N the most abundant nutrient in the plant. It regulates many metabolic functions like the osmotic adjustment, which has an important role in frost and water stress resistance. In spite of that, few studies have evidenced the effect of tissue K on transplanting performance (Christersson 1976).

Non-structural carbohydrates (TNC) comprise starch and a variety of soluble sugars, these latter having a prominent role in cold hardiness and drought tolerance of plants. TNC support respiration and growth especially when photosynthesis is low. Resprouting depends on TNC and in many deciduous species growth of new organs in spring relies on TNC (Dickson and Tomlinson 1996, McPherson and Williams 1998). TNC have proved to be important for transplanting performance in cold-stored seedlings when they deplete their TNC during storage and have low photosynthesis after transplanting (Puttonen 1986). We believe that poor transplanting performance in Mediterranean plantations due to low seedling TNC is probably not a common problem as seedlings rarely are cold-stored, container stock usually has lower transplanting shock than bareroot stock and most species maintain relatively high photosynthetic rates during winter (Villar-Salvador unpublished data).

Fine Root Electrolyte Leakage (REL)

A healthy and vigorous root system is essential for seedling establishment, especially in Mediterranean-climate regions, where seedlings need to extend rapidly their roots before the

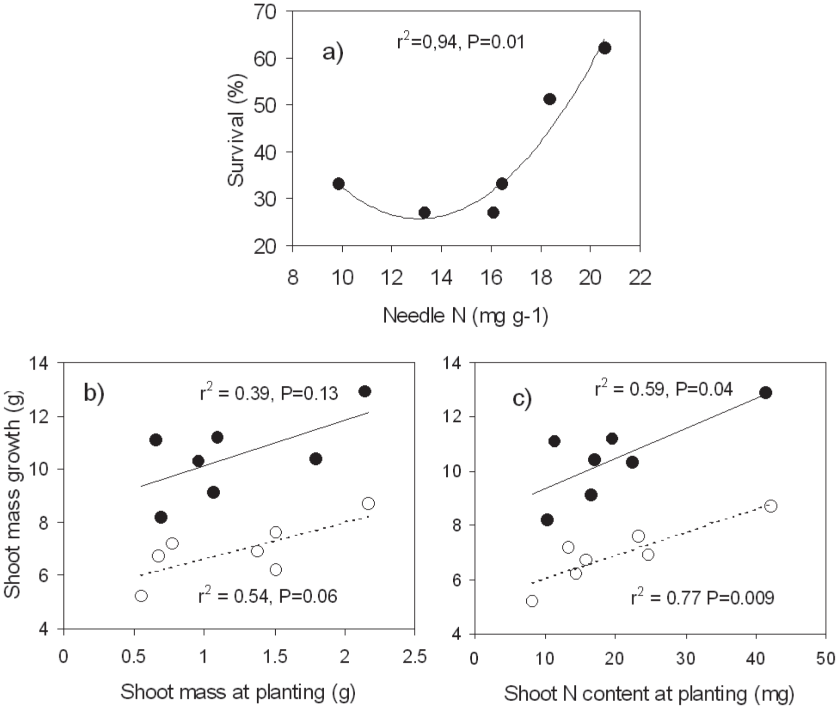

FIGURE 2. (a) Relationship between first year survival and foliar N concentration at planting in *Pinus halepensis* seedlings planted in a semiarid area in SE Spain (Modified from Oliet et al. 1997). Relationships between shoot mass growth two years after planting and shoot mass (b) and shoot nitrogen content (c) before planting in *Pinus halepensis* in two sites of contrasting stress conditions. Solid circles are results in an abandoned field with deep soils (mild stress site), and open circles are results in a slope with stony and shallow soils (high stress site) (Modified from Puértolas et al. 2003b).

onset of summer drought. McKay (1992) described a procedure to assess the vitality of fine roots based on the integrity of cellular membranes. Frost and desiccation can damage cell membrane causing the release ions outside the cell (electrolyte leakage). Electrolyte leakage usually is proportional to membrane damage and to stress intensity and it correlates well with seedling out-planting performance (McKay and White 1997). Electrolyte leakage can also be used to measure the plant's dormancy status. Root electrolyte leakage (REL) is used in plant quality control in the United Kingdom. Its methodology is simple and results can be obtained within 2 days.

Roots of containerised seedlings are sensitive to heavy frosts but damage is typically only detected after planting. In these cases REL may be a promising tool for early detection of frost-damaged plants in nurseries located in cold winter areas.

Performance attributes

Performance attributes are assessed by subjecting whole seedling to certain environmental conditions (optimal or not) and their growth, survival or any other physiological response is evaluated. The most frequently used performance tests are root growth capacity and frost hardiness (Dunsworth 1997).

Root Growth Potential Test (RGP)

It is the ability of a seedling to initiate and elongate new roots within a certain period of time (Ritchie 1985). A simple way to perform this test is to transplant seedlings to larger containers with peat, sand or perlite and placed in an optimum growing environment (wet and warm). This performance test has been used worldwide to assess plant quality due to its simplicity and because it measures the functional integrity and vigour of seedlings. Lots with damaged plants can be detected with this test (Fig. 3). RGP not only depends on the root physiological status, but also on the functional characteristics of the rest of the plant. For instance, RGP has been positively related with N concentration, seedling size, and frost resistance (Ritchie 1985, van den Driessche 1992, Pardos et al. 2003, Villar-Salvador et al. 2004).

From an operational point of view, RGP has two important disadvantages. On the one hand, RGP has important seasonal variations and it may vary depending on previous climatic conditions (Fernández and Pardos 1995). On the other hand, RPG test takes at least one week to be completed.

Although controversial, RGP is used to predict out-planting performance (see Simpson and Ritchie 1997). RGP tends to predict better absolute growth than survival (Fig. 3). Relationships between survival and RGP are frequently asymptotic, indicating that under a specific limit survival diminishes because seedlings are damaged or have low vigour. RGP predicts well the field survival and growth of seedlings in harsh sites where performances is tightly related to seedling vigour (Simpson and Ritchie 1997).

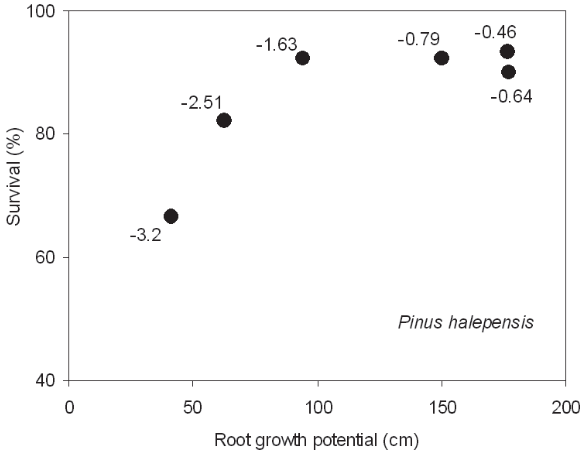

FIGURE 3. Relationship between out-planting survival and root growth potential in *Pinus halepensis*. Each point represents a treatment that experienced different pre-planting drought. For each point, predawn water potential (MPa) of each treatment before planting is represented. (Modified from Vallas-Cuesta et al. 1999).

RGP also predicts growth of vigorous seedlings in mild sites. In other cases, RGP may have limited predictive capacity. To increase the transplanting performance predictive capacity of RGP, some authors have proposed to carry out RGP tests under suboptimal conditions, similar to those that seedlings would encounter when planted (Folk and Grossnickle 1997).

Frost hardiness

Plants from temperate regions acclimate to frost during late summer and autumn. This process is called frost hardening or cold hardening and involves a number of biochemical and physiological changes, which allows plants to avoid freezing injury. Frost hardiness can be assessed subjecting whole seedlings or parts of them to artificial frost and evaluating frost damage. Temperature is reduced at a fixed speed to the target temperature. Cooling rate can determine the degree of injury (Sutinen et al. 2001), so it must be carefully controlled and the same rate used in the different tests. Frost damage can be evaluated at a unique freezing temperature or calculating the lethal temperature at which 50% of the seedlings are killed after subjecting different batches of seedlings to three or four decreasing freezing temperatures. Visual assessment and electrolyte leakage are the most common methods for evaluating frost damage. Visual assessment is simple and quantifies leaf and stem cambium necrosis or seedling mortality. However, it takes several weeks to be completed and requires a greenhouse or a growth chamber to maintain the seedlings under good environmental conditions after freezing. This limits their use for operational plant quality control, but it is a good option for research. The principles and procedures for electrolyte leakage determination are the same described for REL tests and its main advantage over visual damage assessment is its rapidity. However, before using electrolyte leakage in operational plant quality assessment it has to be calibrated for each species with visual frost damage determinations.

Frost hardening of forest species has been extensively studied in boreal and wet-temperate species. In cold climates, it is essential to have information about hardening and dehardening cycles of plants and how the nursery practices and environment influence this processes. In Mediterranean climates, where the main limitation for plant establishment is summer drought, researchers have focused more on plant water relations. However, cold is also important for plant life in Mediterranean climates, especially in inland and highland areas where frosts can last five or six months. There are few studies on frost hardiness of the Mediterranean forest species, yet the importance of low temperature on plant growth, survival, and drought hardiness has been demonstrated in *Q. ilex* and *P. halepensis* (Larcher and Mair 1969, Pardos et al. 2003, Mollá et al. 2006).

Factors that affect plant quality

Plant quality and out-planting performance depends on the environmental conditions and cultural practices in the nursery and how plant is handled after lifting and prior planting. In this section we briefly review these factors.

Fertilization

Fertilization in the nursery has a strong influence on plant morphology and physiology and its transplanting performance. Nitrogen is the most important nutrient and high N fertilization rates increase seedling growth, S/R, photosynthetic rate, N concentration and root growth capacity. Contrary to what many foresters and nurserymen assume, poor fertilization in Mediterranean species usually reduces their transplanting survival and growth (Oliet et al. 1997, Villar-Salvador 2004, Oliet et al. 2005). At present, we do not know yet which is the optimal tissue nutrient concentration for Mediterranean species. Good transplanting performance is obtained when N fertilization rate is greater than 70 mg plant^{-1}. However, plasticity of functional attributes in response to N fertilization is species-dependent, Mediterranean pines being more plastic than oaks (Oliet et al. 1997, Luis et al. 2003, Villar-Salvador et al. 2004).

The ideal amount of fertilizer to grow a seedling is that which maximises seedling nutrient loading and stress resistance without strong morphological imbalances. Excessive N fertilization may reduce frost and drought hardiness and transplanting performance (van den Driessche 1988, Colombo et al. 2001). In Mediterranean species, high N fertilization reduces frost hardiness in *Pinus halepensis* and *P. pinea*, but no effect was observed in *Juniperus thurifera* and *Quercus coccifera* (Puértolas et al. 2005, Villar-Salvador et al. 2005).

Some techniques have been developed to promote nutrient loading of cultivated seedlings without morphological imbalances. One of these techniques consists of maintaining or increasing fertilization after growth cessation in the autumn rather than restricting it, as it is commonly done. Nutrients provided during this period are not diluted with current growth and concentrate in the plant. In Mediterranean pines, late-season N fertilization increased field performance in *P. halepensis* (Puértolas et al. 2003a). However application of late-season fertilization in Mediterranean nurseries located in mild winter areas may be difficult as many species keep growing actively up to the end of the autumn. Exponential fertilization can be a more promising technique for nutrient loading. It is based on the “steady-state nutrition” concept (Ingestad and Lund 1986) and consists of the addition of high fertilizer inputs at exponential rather than constant rates, following seedling growth (Timmer and Aidelbaum 1996). There have been few experiments carried out on Mediterranean species (Carrasco et al. 2001) and much research on this subject is needed before it can be widely recommended.

Irrigation

Water is essential for many basic physiological processes of plants (Landis et al. 1989). Low watering reduces plant growth and nutrient uptake, and increases S/R. It can also cause upgrowing roots if the lower part of the plug remains dry, and salinization of the growing medium. On the contrary, water excess can induce loss of root vitality, proliferation of fungi diseases spread, impairment of root architecture and nutrient leakage. Watering regime must be adjusted through the cultivation and between species. Broadleaved species need special attention due to the high interception of water by leaves, which cause its irregular distribution among plants. In hot climates adequate irrigation is essential to prevent foliage overheating and consequently seedling damage.

Reduction of water supply during the late stages of nursery cultivation has been used to acclimate seedlings to water stress (drought hardening). Drought hardening increases the water stress resistance of plants but the type and magnitude of the response is species-dependent. In Mediterranean species, no clear trend of drought conditioning in the nursery on transplanting performance could be concluded (Vilagrosa et al. 2006).

Growing medium

The function of growing medium is to store water and nutrients that the plant can uptake and to anchor and maintain up straight the plant in the container. Therefore, growing medium has a profound influence on seedling morphology and physiology (Guehl et al. 1989). Peat is the most commonly used growing medium due to physical and chemical properties. Mixtures of this material with other organic and inorganic compounds (sand, perlite, vermiculite, pine bark, etc) are frequently used to improve its structural stability. One of the limitations of peat management is that it becomes hydrophobic if it dries excessively. This property has been considered to hinder seedling establishment in dry sites, although there are no studies that support this. Other growing medium alternatives like coconut fibre, ground pine cones and bark, or saw dust and wine distillery wastes have been tested (Landis et al. 1990). As peat has to be imported in most Mediterranean countries these alternative should be seriously considered, but studies about the effect of different growing media on Mediterranean species are almost inexistent (see Ruano et al. 2001).

Containers

Plant morphology depends on the dimensions of the container used and on cultivation density. Plant size tends to increase with container volume without great effects on S/R. High growing density increases plant height and S/R but reduces root collar diameter, plant mass and the number of lateral branches (Landis et al. 1990, Domínguez-Lerena et al. 2006). Tissue nutrient concentration increased with container volume in *P. pinea* and in general transplanting survival and growth in Mediterranean species tends to be higher when cultivated in large rather than in small volume containers (Domínguez-Lerena 1999, Domínguez-Lerena et al. 2006). Most Spanish nurseries grow Mediterranean species in containers with volumes larger than 200 ml. Stock cultivated in container volumes of 250-300 ml tend to have good transplanting results at an acceptable cultivation costs. Transplanting survival of species with strong taproots, such as oaks, increases when cultivated in deep containers than in shallow containers (Domínguez-Lerena 1999).

Root architecture before and after transplanting is also influenced by container structure. Containers may induce spiralized roots that may reduce out-planting performance and the future structural stability of trees due to poor lateral root egress (Lindström and Rune 1999). This can be prevented by cultivating seedlings in square shape containers or in containers with vertical internal ribs. Although they prevent root spiralling, vertical ribs cause a typical root conformation where lateral roots are forced to grow downward and are air-pruned at the drainage hole. As a consequence, the growth of new lateral roots after planting is concentrated in the lower part of the plug and the root system is far from natural, which may impair tree stability (Rune 2003). A solution is to prune lateral roots. This can be done by chemicals such as copper salts coated in the inside lateral walls of the container or by lateral air pruning, where lateral root tips contact air in vertical slits in the container wall. However this latter system is difficult to implement in nurseries located in hot places because desiccation of growing media is fast and plants need to be watered very frequently.

Seedling storage, rough handling, and nursery location

Seedling quality can be impaired during their storage prior to planting due to seedling desiccation, loss of sugar reserves, and mould development (McKay 1997). Desiccation is more likely to occur in bare-root stock than in containerised stock because the water stored in the plug buffers for desiccation. Therefore it is important to ensure the complete plug is well hydrated at lifting. As most Mediterranean species are cultivated in containers, plant desiccation is not a frequent problem. However, desiccation can occur if plants are stored for prolonged periods at the planting site. *Pinus halepensis* seedlings reduced their transplanting performance when plants desiccated to predawn water potential < -2MPa (Vallas-Cuesta 1999). Prolonged storage of plants in darkness, even at low temperature, can reduce carbohydrate reserves and this can impair transplanting performance (Puttonen 1986). Plant storage in darkness and at low temperature is not a frequent practice in Mediterranean countries because winter is not as cold as at higher latitudes, planting is concentrated in the fall and winter, and it increases cultivation costs.

Plants can be damaged due to rough handling during lifting, transport and planting. Particular care must be taken when plants are loaded and unloaded from vehicles and distributed in the field (McKay 1997).

Finally, nursery location can affect plant quality due to differences in winter temperature among nurseries. Seedlings that are cultivated in nurseries located at higher altitude and latitude are more cold-resistant and harden earlier than seedlings cultivated in nurseries placed at coastal or low latitude sites (Mollá et al. 2006). This is important if stock has to be planted in cold sites.

Concluding remarks

Utilisation of high-quality seedlings is important for revegetation success. Cultivation conditions in the nursery and plant care before planting determine the quality of planted seedlings. Plant quality can be assessed by simple morphological attributes that measure the structure, colour and appearance of the plant. However, morphological attributes have limited capacity to discriminate poor quality plants, being recommendable to complement them by assessing physiological attributes relevant for plant establishment and/or assessing the performance of seedlings to specific environmental conditions.

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