



NURSERY PRE-CONDITIONING OF PLANTS FOR REVEGETATION, GARDENING AND LANDSCAPING IN SEMI-ARID ENVIRONMENTS

J. A. Franco*, J. J. Martínez-Sánchez, J. A. Fernández, S. Bañón, J. Ochoa and M. J. Vicente

Departamento de Producción Vegetal, Unidad Asociada al CSIC de "Horticultura Sostenible en Zonas Áridas" (UPCT-CEBAS), Universidad Politécnica de Cartagena, Paseo Alfonso XIII, 48, 30203 Cartagena, Spain

*Corresponding author, to whom more information request should be addressed (e-mail: josea.franco@upct.es).

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INTRODUCTION



Fig. 1. Drip-irrigated *Asteriscus maritimus* plants in a xerogarden

In landscaping and xerogardening projects, under semi-arid conditions, appropriate techniques used in the nursery during seedling production are crucial for the establishment, survival and subsequent growth of plants after transplanting (Figure 1). Morphological and anatomical adaptations in seedlings include reductions in shoot height and/or leaf area, rises in root-collar diameter and root growth potential and, often, a reduction in the shoot:root ratio; in addition, there are physiological characteristics of seedlings related to osmotic adjustment and water-use efficiency, such as low stomatal conductance, leaf water potential, leaf turgor potential and relative water content. These occur as a result of hardening and acclimation processes (pre-conditioning) during the nursery period, and are correlated with the ability to withstand the shock of transplantation and to increase survival and plant growth following transplantation in xerogardens and semi-arid landscapes (Franco *et al.*, 2006).

Deficit irrigation is the most commonly used pre-conditioning technique to produce high-quality seedlings (Arreola *et al.*, 2006; Franco *et al.*, 2008). In addition, using large-sized containers and appropriate substrates, withholding N nutrition, inoculating arbuscular mycorrhizal fungi, applying plant growth retardants and mechanical conditioning methods are common. Varying microclimatic conditions are also used to control growth to produce high-quality seedlings (Franco *et al.*, 2006).

IRRIGATION MANAGEMENT

Monitoring nursery moisture regimes (Figure 2) and understanding morphological and physiological shoot and root responses of seedlings to water management throughout the nursery period are critical to optimise production of high-quality seedlings (Franco *et al.*, 2006).

Deficit-irrigation during nursery production affected some morphological and physiological aspects related to the hardening of seedlings, or plantlets, of ornamental plants of interest for xerogardening, revegetation and landscaping, plants such as *Limonium cossonianum* (Franco *et al.*, 2002b), *Lonicera implexa* (Navarro *et al.*, 2008), *Lotus creticus* (Franco *et al.*, 2001; Bañón *et al.*, 2004), *Myrtus communis* (Bañón *et al.*, 2002), *Nerium oleander* (Bañón *et al.*, 2005b), *Olea europaea* var. *sylvestris* (Bañón *et al.*, 2003b), *Phillyrea angustifolia* (Fernández *et al.*, 2006), *Rhamnus alaternus* (Bañón *et al.*, 2003c) and *Rosmarinus officinalis* (Sánchez-Blanco *et al.*, 2004). Responses described for different periods of drought depended on the environmental conditions and the species in a study with Mediterranean shrubs (*Pittosporum tobira*, *Callistemon citrinus* and *Rhamnus alaternus*) under water stress conditions in winter, spring and early summer (Miralles *et al.*, 2010).

Deficit-irrigation during the nursery phase reduced the growth parameters of the aerial parts (i.e., plant height, shoot length, leaf area, stem diameter, shoot fresh and dray weight) and root growth (i.e., root length, dray weight and volume) of *Lonicera implexa* (Navarro *et al.*, 2008), *Lotus creticus* (Franco *et al.*, 2001; Bañón *et al.*, 2004), *Myrtus communis* (Bañón *et al.*, 2002), *Nerium oleander* (Bañón *et al.*, 2005b) and *Rosmarinus officinalis* plants (Sánchez-Blanco *et al.*, 2004).

In addition, deficit-irrigation during the nursery phase increased the percentage of thick roots, and reduced the percentage of medium and fine roots in *Myrtus communis* and *Nerium oleander* plants (Bañón *et al.*, 2002; 2005b). Proportionally, root volume was reduced more than dray weight, with the result that root density increased under deficit-irrigation. The hardening of roots, as revealed by an increased percentage of brown roots at lower irrigation rates in *Lotus creticus* (Franco *et al.*, 2001) and *Limonium cossonianum* (Franco *et al.*, 2002b), is of great interest to produce seedlings



that are better adapted to drought-stress at transplantation. The change in colour from white to brown is associated with suberisation of the exodermis, and may reflect a metacutisation process. This is a process of lignification and suberisation that results in a resting root being protected against significant fluctuations in environmental conditions such as drought, and being capable of regrowth when conditions ameliorate. The cortex thickness:root radius ratio was increased in deficit-irrigated *Silene vulgaris* seedlings, increasing the capacity of the plants to obtain water and survive adverse conditions after transplanting (Franco *et al.*, 2008).



Fig. 2. Monitoring *Arbutus unedo* plants using Time Domain Reflectometry (TDR)

At the end of the nursery period, the stomatal conductance (g_s), leaf water potential (Ψ_l), leaf turgor potential (Ψ_p), leaf osmotic potential at full turgor (Ψ_{os}), and relative water content (RWC) of *Nerium oleander*, *Rosmarinus officinalis* and *Myrtus communis* plants grown with deficit-irrigation were lower than those of normally-irrigated plants (Bañón *et al.*, 2002; 2005b; Sánchez-Blanco *et al.*, 2004). Deficit-irrigation produced *Lotus creticus* seedlings with greater leaf trichome densities and numbers of xylem vessels in stems and roots, and induced a range of physiological adaptations. In addition, seedlings undergo osmotic adjustment, which might provide considerable capacity to adapt to adverse conditions during the field establishment period (Franco *et al.*, 2002a; Bañón *et al.*, 2004). Also, *Phillyrea angustifolia* plants watered at 40% water-holding capacity throughout the nursery phase showed higher stomatal density and water-use efficiency, which would allow the plants to overcome transplantation shock (Fernández *et al.*, 2004).

Deficit-irrigation of *Nerium oleander* (Bañón *et al.*, 2005b) seedlings during nursery production clearly reduced their mortality rates after transplantation under drought and heat conditions. Similarly, seedlings of *Lotus creticus* (Franco *et al.*, 2001; 2002a) and *Limonium cossonianum* (Franco *et al.*, 2002b) that were watered less in the nursery showed greater and faster root development following transplantation under semi-arid conditions, especially when soil moisture was low (Franco *et al.*, 1999). This adaptive potential of plants perseveres, and a small amount of water after a long period of drought reactivated root growth more rapidly in *Limonium cossonianum* plants that had been less watered during the nursery period (Franco *et al.*, 2002b).

On the other hand, the irrigation system can influence root growth, resulting in differences in lateral and basal root elongation (Franco and Leskovar, 2002).

APPLICATION OF PLANT GROWTH REGULATORS

Paclobutrazol (PBZ), a growth retardant and member of the triazole group, induces mild water-stress tolerance and can increase water-use efficiency in seedlings and adult plants (Franco *et al.*, 2006). PBZ reduced plant height and shoot and root dry weight in *Nerium oleander* seedlings (Ochoa *et al.*, 2009). PBZ-induced drought-tolerance has been associated with decreases in transpiration, plant height, biomass and leaf area, and with an increase in stomatal resistance in nursery-produced *Phillyrea angustifolia* seedlings (Fernández *et al.*, 2004). These effects increased the percentage of plants that survive after transplantation into semi-arid conditions (Bañón *et al.*, 2001b). Similarly, PBZ reduced plant height and aerial dry weight, increased root diameter and root system volume, and reduced stomatal conductance in *Arbutus unedo* (Navarro *et al.*, 2004).

PBZ reduced the symptoms of saline-stress in *Nerium oleander* seedlings by reducing the uptake and accumulation of harmful Na^+ and Cl^- ions by plant tissues, and promoted a process of osmotic adjustment through the accumulation of organic osmotic compounds (Bañón *et al.*, 2005a). Also, PBZ reduced the symptoms of saline-stress and mortality in *Rhamnus alaternus* seedlings in different ways; by increasing stomatal conductance, by promoting organic solute synthesis and by reducing the availability of salt ions in the medium (Bañón *et al.*, 2003d).

Chlormequat chloride (CCC) reduced the dry weight of the aerial parts of *Nerium oleander* plants grown in a nursery, thereby producing plants with a darker and duller colour (Bañón *et al.*, 2001a). Similarly, both PBZ and ethephon (ETH) controlled stem height in *Reichardia tingitana* and improved its ornamental value (Bañón *et al.*, 2003a).

The combined effect of deficit-irrigation and PBZ during the nursery phase also improved the resistance mechanisms related to the plant adaptation to drought conditions in *Lonicera implexa* (Navarro *et al.*, 2008)

PRUNING

Frequently during nursery production, air or chemically root-pruned seedlings *Nerium oleander* plants in the nursery produced shorter plants with more shoots, greater root length and lower shoot:root ratios (Bañón *et al.*, 2001a).



TEMPERATURE MANAGEMENT

Modification of nursery temperature regimes can influence the performance of nursery stock after transplantation into the field. Lowered temperatures reduced shoot length and the shoot:root ratio, and increased the percentage of brown roots in *Lotus creticus* (Franco *et al.*, 2001). Low temperatures increased Ψ_l and the abaxial stomatal density of water-stressed *Lotus creticus* plants (Bañón *et al.*, 2004).

Low nursery temperatures continue to affect growth after planting, increasing relative growth rates. This effect was found in *Lotus creticus*, with plants from an unheated greenhouse showing slightly higher root growth than plants from a heated greenhouse after transplanting with different levels of establishment irrigation (Franco *et al.*, 2001).

When the combined effects of deficit-irrigation and low temperature during the nursery period are studied (Franco *et al.*, 2001; 2002a; Bañón *et al.*, 2004), regimes involving the least water and lowest temperatures produce plants best adapted to stress during transplantation. These plants have lower shoot:root ratios, lower shoot fresh weight:length ratios, higher percentages of brown roots, lower g_s , Ψ_l , Ψ_p , and RWC values, higher leaf trichome densities and greater numbers of xylem vessels in stems and roots. The most stressed seedlings in the nursery (i.e., those that received least water and low temperatures) show greater and more rapid root growth after transplantation, especially when soil moisture content is low.

MANAGEMENT OF AIR HUMIDITY

Bañón *et al.* (2003c), working with *Rhamnus alaternus*, reported that low air humidity and high water-deficit during the nursery phase reduced shoot growth, Ψ_l , leaf g_s and net CO₂ assimilation rate, leading to improving the survival of seedlings at the end of the establishment period. The effect of low air humidity on Ψ_l of *Olea europaea* var. *sylvestris* was less marked than the effect of deficit-irrigation. However, Ψ_p values were higher, suggesting that low air humidity pre-conditioning might trigger an elastic adjustment (this mechanism enhances cell wall elasticity and avoids any substantial reduction in the leaf turgor) in this species (Bañón *et al.*, 2003b). The combined effect of both hardening treatments increased the rate of survival after transplantation by around 63% compared with the control (0%).

In a subsequent study, the combined effect of deficit-irrigation and low air humidity during the nursery phase also reduced the mortality rate of *Nerium oleander* seedlings after transplantation under drought and heat conditions compared with controls (from 93% to 32%; Bañón *et al.*, 2005b). Similarly, the above-mentioned combined effect reduced the mortality rate of *Myrtus communis* seedlings after transplantation under drought and heat conditions from 100% to 66%, compared with the controls (Bañón *et al.*, 2002). Such behaviour was related to morphological changes observed in the aerial parts (i.e., smaller plant size and lower leaf area), in the roots (i.e., shorter, thicker, more dense and less ramified) and in the shoot:root ratio (i.e., reduced by approx. 60% in both species) of the pre-conditioned plants.

IRRIGATION FOLLOWING TRANSPLANTING

Commonly, seedlings of different species irrigated frequently following transplantation generate a greater new root mass and establish more quickly than those that receive infrequent irrigation, although there are wide variations in this response (Franco *et al.*, 2006). Thus, root development of *Limonium cossonianum* plants was substantially lower when there was no establishment irrigation than when a single establishment irrigation of 50 l m⁻² was applied in semi-arid conditions (Franco *et al.*, 2002b). However, establishment of plants in xerogardens or in arid and semi-arid landscapes, where available irrigation water is limited, is often costly and problematic.

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