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Testing Nursery Plant Quality of Canary Island Pine Seedlings Grown under Different Cultivation Methods

By

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Summary

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This study tests the influence of different nursery practices, which may affect final plant quality. Different containers, natural and artificial substrates, and concentration of slow-release fertilizers were tested to determine their influence on morphological, physiological and performance attributes of *Pinus canariensis*, a species of great forestry interest, endemic in the Canary Islands.

Physiological attributes were important in order to know that in all treatments presented nutrient and non-structural carbohydrates concentrations are within or near the optimal ranges described for conifers, but there are no differences between them. The applied conditions in The Oregon State University test (OSU) had to be modified for this species because the stress conditions (40°C during 50 min) were not enough to provoke seedlings death. As for the morphological attributes; seedling height and the Dickson Quality Index (DQI) were the most informative parameters. These parameters together with the root growth potential test (RGP), showed significantly better results for plants grown in artificial substrates than natural ones and, particularly, for those grown in Arnabat containers.

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Zusammenfassung

LUIS V. C., PETERS J., GONZÁLEZ-RODRÍGUEZ A. M., JIMÉNEZ M. S. & MORALES D. 2004. Überprüfung der Baumschulqualität von unter verschiedenen Methoden kultivierten Kanaren-Kiefer-Sämlingen. – Phyton (Horn, Austria) 44 (2): 231–244. – Englisch mit deutscher Zusammenfassung.

In dieser Untersuchung werden verschiedene Anzuchtmethoden, welche die spätere Pflanzenqualität beeinflussen könnten, überprüft. Verschiedene Container, natürliche und künstliche Substrate sowie die Konzentration langsam wirkender Dünger werden getestet, um deren Einfluss auf morphologische, physiologische und leistungsmäßige Merkmale von *Pinus canariensis* zu bestimmen. Diese Species ist endemisch auf den Kanaren und von großem forstlichen Interesse.

Die physiologischen Merkmale sind deshalb wichtig, da man aus ihnen ersehen kann, dass die Nährstoff- und die nicht strukturgebundenen Kohlenhydratkonzentrationen in allen Behandlungen innerhalb oder nahe dem optimalen Bereich für Koniferen liegen. Innerhalb der einzelnen Behandlungen gibt es keine Unterschiede. Die Bedingungen für den „The Oregon State University Test (OSU)" mussten für diese Baumart modifiziert werden, da Stressbedingungen (40° C während 50 min.) nicht ausreichten, um den Tod von Sämlingen hervor zu rufen. Zu den morphologischen Kennzeichen: Die Höhe der Sämlinge und der „Dickson Quality Index IDQ)" sind die aussagekräftigsten Parameter. Diese sowie der Test auf das Wurzelwachstum (RGP) zeigen signifikant bessere Ergebnisse bei Pflanzen, welche in künstlichen Substraten gegenüber natürlichen wuchsen und zum Teil auch bei solchen, die in Arnabat- Containern aufgezogen wurden.

Introduction

Successful establishment of plants for reforestation depends upon a wide range of interacting factors including: climate, soil, competing species and post-plantation care. Plant quality is one aspect that is increasingly recognized as contributing to good plant establishment (SOUTH 2000, PUÉRTOLAS & al. 2003). Therefore, for successful seedling establishment in the field, it is necessary to understand how nursery practices affect seedling quality, which in turn is influenced by a complex system of plant morphological and physiological conditions and which is studied through quality attributes.

Over the last 50 years, and especially during 80s and 90s, scientists and foresters studied ways to produce better seedlings through improved nursery culture, and developed tests to assess seedling quality as those based in morphological parameters, nutrient and carbohydrate analysis and performance test (DURYEA 1985, SOUTH 2000, MASON 2001). The achievement of better cultivation practices and higher survival rates has erroneously given rise to the impression that stock quality assessment is a tool of the past. The high cost of reforestation and low percentages of survival at some sites make these kinds of studies very important, especially when beginning to reforest with species not studied in this respect before. There are many studies describing the best conditions to obtain good quality plants for reforestation in different pine species but, up to now, this has not been carried out for the Canary Island pine.

Pinus canariensis C. Sm. ex DC. in Buch is an endemic species of the Canarian Archipelago (Spain), a relict of the Tertiary Thetys vegetation (PAGE 1974). It forms the pine forests existing on the highest islands, and has been planted all around the world due to its high ornamental and forestry value as well as fire resistance. Due to massive exploitation during the conquest of the islands in the past, this pine forest has been reduced to just 13% in Tenerife (DEL ARCO & al. 1992). From this remaining nucleus the Spanish authorities began a large reforestation and plantation campaign in Tenerife in the 1940s, which led to an almost complete belt of pine forest around the highest summits of this island. Nevertheless, during the last 20 years there have been problems related to the natural regeneration of pine seedlings in some areas of Tenerife (LUIS & al. 2001) and at present reforestation has started again in Tenerife in order to complete this belt. Good quality planting stock is an essential precondition for reforestation success and will contribute to better results when this species is planted in other parts of the world.

The aim of this study was to test the influence of different nursery cultivation practices (mainly types of containers, substrates and fertilization) on final nursery plant quality of *P. canariensis* seedlings for reforestation, using morphological, physiological and performance test.

Material and Methods

Plant Material and Cultivation

Pinus canariensis seeds from Vilaflor (Tenerife, Canary Islands, Spain) provenance (FS-27/01/38/004) were soaked for 24 hours in a water bath to discard empty seeds by flotation. Afterwards, they were sown in two types of commercial containers in mid-June 2001. There were 4 different substrate mixtures and three fertilization treatments (see Table 1). We distinguished between natural substrates (treatments 1, 6, 7 and 12) which contained soil in the mixture, and artificial substrates, which had no soil (treatments 2, 3, 4, 5, 8, 9, 10 and 11). The following materials were used: Containers: a) Super Leach M-30 (SL). Cavity volume 305 ml. Individual cells (in a tray of 35 cells). Depth 17.5 cm. Density 262 seedling m^{-2} (Steuwe & Sons Inc., Corvallis, USA); b) Arnabat 48C (AT). Cavity volume 308 ml. 48 cells in a compact container. Depth 17.4 cm. Density 378 seedling m-2 (Arnabat S.A., Spain). Substrates: a) Soil: Soil from the natural pine forests of Tenerife; b) Lapilli: volcanic material very commonly used in the Canary Islands for agriculture, which maintains humidity and helps ventilation to the substrates; c) Peat: Floratorf® (Floragard Product, Germany); d) Vermiculite: Eurover Vermiculita (Europerlita S.A., Spain). Fertilizer: It is a commercial slow-release fertilizer called Osmocote Plus: 16/ 8/12 (N/P/K) (Scotts, The Netherlands) in rates of i) none; ii) 4 g l^{-1}; iii) 7 g l^{-1}. Plants were raised in the nursery of the Centro Ambiental La Tahonilla Baja in La Laguna (Tenerife). Ten containers per treatment were sown and placed randomly. Seedlings were well watered depending on substrate mixtures, climate and plant appearance, following the nursery personnel criteria. After 8 months of growth, morphological, physiological and performance attributes were analysed.

Morphological Attributes

Twenty five seedlings per treatment were randomly selected to measured Shoot height (H) in cm; root collar diameter (D) in mm, (measured with a digital micrometer, Mitutoyo Corporation. APB-1D. 1PC. Japan); Sturdiness Quotient (SQ), which relates height (cm) and root collar diameter (mm) [SQ = H/D]; shoot and root dry weight (DWs and DWr, respectively); shoot: root dry weight ratio [DWs/DWr]; Dickson Quality Index (DQI), calculated from other analysed attributes [DQI = DWt/(SQ+DWs/DWr)] where DWt is total dry weight; and seedling extraction ease (SEE), evaluated by a numerical classification: 1 Minimum ease: it is necessary to employ an object, pushing from the base of the container, to remove the seedling and, as a consequence, damage to the seedling occurs, 2 Medium ease: Complications during the extraction occurs, but finally the seedling can be removed from containers without damage, 3 Greatest ease: Seedling very easily removed and no damage occurs.

Table 1

Composition of the 12 different treatments studied (1-12). Soil (S); peat (P); Lapilli (L); Vermiculite (V) and Osmocote fertilizer (Os), fertilization rate: 4 g l^{-1} and 7 g l^{-1}.

Containers	Substrates	Fertilization	Treatment
Super Leach M-30 (SL)	2/3 S + 1/3 L	No	1
	2/3 P + 1/3 V	Os 4 g l^{-1}	2
		Os 7 g l^{-1}	3
	2/3 P + 1/3 L	Os 4 g l^{-1}	4
		Os 7 g l^{-1}	5
	1/3 S + 1/3 P + 1/3 L	No	6
Arnabat 48C (AT)	2/3 S + 1/3 L	No	7
	2/3 P+ 1/3 V	Os 4 g l^{-1}	8
		Os 7 g l^{-1}	9
	2/3 P+ 1/3 L	Os 4 g l^{-1}	10
		Os 7 g l^{-1}	11
	1/3 S + 1/3 P + 1/3 L	No	12

Physiological Attributes

After seedlings were removed from containers, ten seedlings per treatment were washed with water and dried at 60 °C. They were ground to 0.7 mm. and the powder was stored in sealed plastic tubes until it was tested under the different protocols. A ground sample (1 g) was used for nitrogen determination in needles (N_{needle}) and roots (N_{root}) according to the Kjeldahl method. Phosphorus concentration in needles (P_{needle}) and roots (P_{root}) was measured by the vanadate-molybdate espectrophotometric method (Peter 1942). Potassium concentration in needles (K_{needle}) and roots (K_{root}) was analyzed by a flame photometer method (Berneking & Schrenk 1957). Soluble concentration sugars (SS) (Irigoyen & al. 1992) and Starch (St) (Rose & al. 1991) was determined by an anthrone spectrophotometric method.

Performance Attributes

The Oregon State University test (OSU) was assessed following McCreary & Duryea 1985 in order to know plant vigor after a temperature stress. Ten seedlings per treatment were exposed to a temperature of 40 °C for 50 min and ten seedlings were taken as a control. Afterwards, seedlings were planted into plastic bags of 300 ml with a mixture 1:1 of peat and vermiculite in order to determine survival after one and two months. During this period plants were well watered in a greenhouse with a photoperiod of 16 h and temperatures between 15°C–25°C.

Root Growth Potential test (RGP) was applied following Ritchie 1985, in order to know the ability of seedlings to initiate and elongate roots. Ten seedlings per treatment were randomly selected. White roots were cut and the seedlings were planted in plastic bags of 2 L containing perlite in a greenhouse with a photoperiod of 16 h and temperatures between 15°C–25°C. One month later the number of new white roots was counted (Nprr) and their length was measured (Lprr) after this time.

Statistical Analysis

Morphological, physiological and performance attributes were compared between sets using two way ANOVA analyses. Tukey's multiple range test was applied when set effect was significant ($\alpha = 0.05$). All statistical tests were performed by using SPSS (v.11.5 SPSS Inc., Chicago, IL, U.S.A).

Results

Morphological Attributes

Table 2 shows morphological parameters for the seedlings of the 12 different treatments. At the end of the cultivation period, maximum height was achieved by seedlings from treatment 8, 9 and 11 and minimum height was achieved by seedlings from treatment 1, 6, 7 and 12. Greatest root collar diameter was achieved in treatments 8, 9 and 11 with no significant differences between them. The sturdiness quotient (SQ) showed significant differences between seedlings grown in natural and artificial substrates, being smaller for those grown in natural substrates. Statistical differences could also be observed in shoot:root dry weight ratio between seedlings grown in natural and artificial substrates. There were significant differences in DQI between containers, this attribute being higher in

AT, with significantly higher values in seedlings in treatments 8 and 11. Table 2 shows seedling extraction ease for all treatments, using the numerical classification defined in the section of Material and Methods. Plants grown in natural substrates presented less extraction ease than in artificial ones.

Physiological Attributes

No physiological attribute showed significant differences between containers, therefore we present the results (Tables 4 and 5) combining data of treatments 1 and 7, 2 and 8, 3 and 9, and so on. On the other hand, we always observed significant differences between nutrient concentrations in needles and roots of the seedlings, and the same for soluble sugar concentrations, so we present separate means and standard errors values for both plant parts. We do not do this for starch concentrations which were not significantly different between needles and roots.

Total Nitrogen. Seedlings grown in natural substrates had lower nitrogen concentrations than seedlings from artificial ones. In general root nitrogen concentrations were significantly lower than in needles. Phosphorus. Higher values were found in treatments 3 and 9. Potassium. The lowest potassium concentrations for needles were found in treatments 3 and 9. Except for treatments 3 and 9, needles presented higher potassium concentration than roots (Table 3).

Table 2

Results of morphological attributes analysis 8 months after sown; Height (H); Root collar diameter (D); Sturdiness quotient (SQ); Dry weight ratio between shoot and root (DWs/DWr); Total dry weight (DWt); Dickson quality index (DQI) and seedling extraction ease (SEE) for all treatments (T: 1–12); Mean and standard errors, column values not followed by the same letter are significantly different ($P < 0.05$).

T	H [cm]	D [mm]	SQ [cm mm^{-1}]	DWs/DWr	DWt [g]	DQI [g mm cm^{-1}]	SEE
1	7.9 ± 1.4^{a}	2.4 ± 0.3^{a}	3.3 ± 0.5^{a}	1.25 ± 0.40^{a}	1.12 ± 0.29^{a}	0.25 ± 0.07^{a}	1
2	16.7 ± 2.7^{b}	3.5 ± 0.6^{b}	4.8 ± 0.7^{bc}	2.30 ± 0.48^{b}	3.49 ± 1.09^{b}	$0.55 \pm 0.18b^{cd}$	3
3	19.7 ± 2.5^{c}	4.0 ± 0.5^{c}	5.0 ± 0.7^{bcd}	2.93 ± 0.6^{cd}	3.34 ± 0.77^{b}	0.43 ± 0.12^{b}	2
4	16.0 ± 3.0^{b}	$3.7 \pm 0.6b^{c}$	4.4 ± 0.6^{b}	2.68 ± 0.64^{bc}	3.56 ± 1.17^{b}	0.50 ± 0.16^{bc}	3
5	20.0 ± 3.8^{c}	4.0 ± 0.7^{cd}	5.1 ± 1.6^{cd}	3.21 ± 0.76^{d}	$4.84 \pm 1.42c$	0.61 ± 0.28^{cde}	3
6	7.7 ± 1.2^{a}	2.2 ± 0.2^{a}	3.5 ± 0.6^{a}	1.32 ± 0.56^{a}	1.01 ± 0.32^{a}	0.22 ± 0.09^{a}	1
7	8.2 ± 1.8^{a}	2.2 ± 0.3^{a}	3.7 ± 0.7^{a}	1.08 ± 0.42^{a}	1.09 ± 0.37^{a}	0.23 ± 0.08^{a}	1
8	24.5 ± 3.8^{d}	$4.4 \pm 0.5d^{e}$	5.6 ± 1.0^{d}	2.34 ± 0.49^{b}	5.70 ± 1.25^{de}	0.73 ± 0.19^{ef}	3
9	25.5 ± 3.1^{d}	4.5 ± 0.5^{e}	5.7 ± 0.6^{d}	2.62 ± 0.58^{bc}	5.48 ± 1.01^{cd}	0.66 ± 0.14^{de}	3
10	16.2 ± 2.3^{b}	3.5 ± 0.7^{b}	4.8 ± 0.9^{bc}	2.39 ± 0.81^{b}	3.22 ± 1.23^{b}	0.47 ± 0.23^{b}	2
11	24.6 ± 3.5^{d}	4.7 ± 0.8^{e}	5.3 ± 0.9^{cd}	2.61 ± 0.59^{bc}	6.37 ± 0.99^{e}	0.81 ± 0.16^{f}	2
12	8.8 ± 1.7^{a}	2.5 ± 0.4^{a}	3.6 ± 0.7^{a}	1.30 ± 0.47^{a}	1.17 ± 0.34^{a}	0.24 ± 0.09^{a}	1

Table 3

Results of the analysis of nutritional parameters 8 months after sown; Nitrogen concentration in needles (N_{needle}); Nitrogen concentration in roots (N_{root}); Phosphorus concentration in needles (P_{needle}); Phosphorus concentration in roots (P_{root}), Potassium concentration in needles (K_{needle}) and potassium concentration in roots (K_{root}); for all treatments (T: 1–12). Since no significant differences were found between containers of respective substrates in previous statistical analysis, results of corresponding treatments are presented together. Mean (mg. g^{-1} DW) and standard errors, column values not followed by the same letter are significantly different ($P < 0.05$).

T	N_{needle} [mg g^{-1}]	N_{root} [mg g^{-1}]	$P_{needles}$ [mg g^{-1}]	P_{root} [mg g^{-1}]	K_{needle} [mg g^{-1}]	K_{root} [mg g^{-1}]
1.7	12.0 ± 0.4^{ab}	6.4 ± 0.1^{a}	2.1 ± 0.3^{c}	1.1 ± 0.2^{a}	6.6 ± 1.2^{ab}	2.6 ± 0.4^{a}
2.8	22.5 ± 0.6^{cd}	14.4 ± 0.2^{b}	1.5 ± 0.4^{b}	1.4 ± 0.7^{a}	8.1 ± 2.8^{bc}	5.1 ± 2.8^{ab}
3.9	17.2 ± 0.5^{bc}	19.9 ± 0.5^{c}	2.2 ± 0.6^{c}	1.9 ± 0.3^{b}	4.5 ± 0.6^{a}	4.8 ± 0.5^{ab}
4.10	23.9 ± 0.3^{d}	12.5 ± 0.4^{b}	0.9 ± 0.3^{a}	0.8 ± 0.5^{a}	9.8 ± 2.0^{c}	6.7 ± 3.8^{b}
5.11	22.6 ± 0.6^{cd}	5.1 ± 0.1^{a}	1.5 ± 0.1^{b}	1.4 ± 0.4^{ab}	5.7 ± 0.9^{a}	2.8 ± 1.4^{a}
6.12	8.0 ± 0.4^{a}	5.6 ± 0.1^{a}	2.8 ± 0.5^{d}	1.0 ± 0.3^{a}	8.2 ± 2.1^{bc}	4.2 ± 1.6^{ab}

Non Structural Carbohydrates (NSC). No clear tendency was observed in soluble sugars (SS) (see Table 4). Maximum values for starch were for treatments 2 and 8, although significant differences were not found.

No significant correlations between the analyzed parameters (both morphological and physiological) neither with performance attributes were found.

Table 4

Results of the analysis of non-structural carbohydrates (Soluble carbohydrates and Starch) 8 months after sown; Soluble sugar concentration in needles (SS_{needle}); Soluble sugar concentration in roots (SS_{root}); Starch concentration in the seedling (St); for all treatments (T: 1–12). Since no significant differences were found between containers of respective substrates in previous statistical analysis, results of corresponding treatments are presented together. Mean and standard errors, column values not followed by the same letter are significantly different ($P < 0.05$).

T	SS_{needle} [mg g^{-1}]	SS_{root} [mg g^{-1}]	St [mg g^{-1}]
1.7	126.4 ± 32.8^{a}	78.8 ± 38.3^{ab}	122.0 ± 80.6^{ab}
2.8	125.6 ± 34.3^{a}	102.0 ± 54.6^{b}	171.5 ± 64.4^{b}
3.9	164.5 ± 43.2^{b}	81.9 ± 49.1^{ab}	112.4 ± 42.6^{a}
4.10	116.7 ± 23.2^{a}	72.2 ± 32.5^{ab}	128.0 ± 57.9^{ab}
5.11	128.3 ± 30.8^{a}	90.0 ± 34.1^{b}	131.4 ± 33.0^{ab}
6.12	102.3 ± 27.5^{a}	51.7 ± 20.5^{a}	125.0 ± 42.7^{ab}

Table 5. Root growth potential test: Total number of new produced roots (Nprr) and total length of new produced roots (Lprr) for the seedlings of all treatments (1-12) after one month of growth in perlita. Means and standard errors. column values not followed by the same letter are significantly different ($P < 0.05$).

Substrates	Nprr	Lprr
1.7	6 ± 10^a	4 ± 6^a
2.8	29 ± 23^b	22 ± 17^b
3.9	35 ± 30^b	26 ± 24^b
4.10	39 ± 27^b	29 ± 18^b
5.11	58 ± 28^c	48 ± 24^c
6.12	4 ± 9^a	2 ± 2^a

Performance Attributes

OSU test. After applying the OSU test for each treatment, none of the seedlings died after one month or even two. All seedlings presented a similar good appearance, except treatments 3 and 9, which presented a certain degree of dryness in their needles, approximately 1:3. This was also observed for the control seedlings from this treatment. Root Growth Potential test (RGP). Significant differences were not observed between containers, so data are presented jointly from treatments as previously mentioned (Table 4). It was observed that natural substrates (1, 6, 7 and 12) achieved the lowest values (almost insignificant compared with artificial substrates) of Nprr (total number of new produced roots) and Lprr (total length of new produced roots). Seedlings from treatment 5 and 11 presented the highest number of new roots produced and also the longest new roots produced.

Discussion

Seedlings grown in artificial substrates and Arnabat containers were, in general, taller and had bigger root collar diameter. Height values for different treatment seedlings were within the range allowed for field transfer of conifers (PEÑUELAS & OCAÑA 2000) with the exception of the seedlings grown in natural substrates which did not reach 10 cm height. Seedlings grown in substrates 8, 9 and 11, which statistically slightly exceeded the recommended 25 cm height, were not considered outside the range because many studies (VILLAR-SALVADOR & al. 2000, PUÉRTOLAS & al. 2003) have found a positive correlation between height and survival. With regard to root collar diameter, all seedlings were acceptable for field transfer (PEÑUELAS & OCAÑA 2000), with a diameter above 2 mm. In the past, small plants have been used for reforestation, because they were thought to be less sensitive to wind, drought and cold stress. However, new studies with *Pinus* and *Quercus* species show that larger plants with a balanced shoot: root dry weight ratio can have equal or better field performance (South 2000, Villar-Salvador & al. 2000, Mason 2001). The sturdiness quotient was high, indicating very slender seedlings which are susceptible to wind, drought and cold, nevertheless, this index can be misleading depending on plant height and therefore, plants with high SQ could produce a worse response in the field, if they are unbalanced (Villar-Salvador & al. 2000). Lower shoot: root ratio means, in general, better balanced seedlings (taking into account photosynthesis, transpiration and water absorption by the roots) because the aerial parts have better water and nutrient availability due to a better developed root system. As to the ratio shoot:root, seedlings grown in natural substrates exhibited lower values than the others, but this is due to their lower weight values of the aerial parts. It is clear that seedlings grown in artificial substrates are quite balanced and present a good root development to maintain the transpiring parts, therefore their potential for field performance will be higher (South 2000, Villar-Salvador & al. 2000). Although seedlings grown in natural substrates (treatments 1, 6, 7 and 12) have the best (lowest) DWs/DWr ratio and the highest sturdiness quotient, these seedlings did not reach either the minimum recommended height, which is the most predictive parameter, or a good Dickson quality index, which is a highly developed morphological index to predict field performance (Dickson & al. 1960). Plants grown in natural substrates presented less extraction ease, so that the extraction caused damage to the seedlings in both containers. Plants grown in artificial substrates were much easier to extract. As to the differences between containers Arnabat (AT) was worse than Super-Leach (SL) due to the fact that cells of these containers can not be manipulated individually, but this was the only parameter that would prefer SL over AT. Extraction ease was medium in Arnabat containers with artificial substrates (10 and 11) because the substrate mixture of these treatments (2/3 Peat + 1/3 Lapilli + Osmocote) was more compact than the other artificial ones.

Physiological attributes have recently been found to provide useful information, but in our case the parameters studied (nutrient status and non-structural carbohydrates) did not show clear tendencies between different treatments. All physiological values were acceptable for field transfer, and although these tests were not able to differentiate between treatments, it was very important to know that the seedlings were in the optimal range for these parameters (Villar-Salvador & al. 1999, Puértolas & al. 2003, Tausz & al. 2004). Seedling nutrient status is more related to the growth after planting in the field than to initial survival, because an adequate and balanced nutrient status provides a reserve of mineral elements for growth of new tissues until the seedling can be established in the field (LANDIS 1985). It has been known for a long time (WAKELEY 1948) that mineral nutrition is just one of a number of physiological characteristics that contribute to a healthy seedling. An adequate mineral nutrient level is no guarantee of vitality, but concentration value of nutrients below the optimal range would suggest a disadvantage for survival and growth after out planting, therefore seedlings should be charged with optimum levels of mineral nutrients when planted (LANDIS 1985). Field performance of conifer species has been positively related to shoot nitrogen concentration (VAN DEN DRIESSCHE 1987, VILLAR-SALVADOR & al. 2000, PUÉRTOLAS & al. 2003). Higher levels of nitrogen help seedlings to grow after transplanting and with levels in needles around 2–2.2 % the survival is the greatest (VAN DEN DRIESSCHE 1987). In our experiment treatments 2, 4, 5, 8, 10 and 11 were in this range. Phosphorus stimulates root growth versus aerial part growth, contributing to the production of a more balanced plant (TIMMER & ARMSTRONG 1987, DOMÍNGUEZ-LERENA & al. 2000, RUÍZ & DOMÍNGUEZ 1997 (unpublished)). Mean values of phosphorus in plants are around 2 mg g-1 DW, and for plants growing in containers between 2–4 mg g^{-1} DW (PEÑUELAS & OCAÑA 2000). In our study some treatments presented values lower than this, but this could be due to the fact that high nitrogen values lead to a dilution effect on other nutrients such as phosphorus (MILLER & TIMMER 1994). Although the best known role of phosphorus in plant physiology is the osmotic adjustment of the stomata opening regulation (TIMMER 1991, VAN STEENIS 1999), it also plays an essential role in seedling establishment. Potassium concentration within the tissues is related to plant vigor. In our study treatments 3 and 9 showed lowest values of potassium and resulted in lower seedling vigor in the OSU test.

A very important physiological attribute for quality and for out planting response is the content of non-structural carbohydrates in roots (PUTTONEN 1986, ROSE 1992). Furthermore, in cases of defoliation and herbivores, the accumulated carbohydrates play a fundamental role in plant recovery (MCPHERSON & WILLIAMS 1998, CANHAM & al. 1999). Therefore, a minimum level of nutrient and non-structural carbohydrate concentration is necessary for successful establishment in the field, although it does not ensure seedling survival. Values of NSC for all treatments were within the optimal range (PEÑUELAS & OCAÑA 2000). In conifers, new root formation and plant maintenance during the first stages in the field is mainly the result of the sugars obtained from photosynthesis and, to a lesser extent, from resource sugars (VAN DEN DRIESSCHE 1987, PHILIPSON 1988). Soluble sugars play an important role in osmotic adjustment (GEBRE & al. 1994, PREMACHANDRA & al. 1995) and in dehydration tolerance processes (SANTARIUS 1973) in shoots and in roots (KOPPENNAL & al. 1991, VILLAR-SALVADOR & al. 1999). VILLAR-SALVADOR & al. 1999 reported, for *Pinus halepensis*, higher values for starch content in roots than in shoots, but although all seedlings were within the range, in our case we did not observe this tendency. A decline in nutrient availability has been associated with starch accumulation (ARIOVICH & CRESSWELL 1983), but no clear tendencies were observed in our experiment.

In general, considering all kinds of attributes, there were no differences between fertilization rates within the range studied, so we recommend the use of the lower fertilization rate used (4 g l^{-1} of Osmocote) for economic reasons.

The OSU test must be optimized for *P. canariensis* seedlings. The fact that no seedling died after the stress treatment suggests that, due to the very good heat resistance of the Canarian Island pine (PETERS & al. 1999), applied temperature and exposure time (40 °C during 50 minutes) were not stressful enough, even being higher than those applied to other pine species which resulted in mortality under less stress (McCREARY & DURYEA 1985). As for morphological attributes, with respect to the RGP test, the best results were obtained in artificial substrates, seedlings grown in natural ones showed very low root regeneration potential. The capacity to produce new roots has been found to be an indicator of plant vigor (SIMPSON & RITCHIE 1997) because it has been positively correlated to survival expectation in the field (GIL & PARDOS 1997). This suggests that these seedlings would present the highest establishment rates in the field. This test is considered superior to many other seedling quality tests because it indicates seedlings resistance to stress and reflects stocklot performance potential (RITCHIE & TANAKA 1990, SUTTON 1990). However, we have to take into account that RGP indicates seedling quality but not site or plantation quality and so, successful performance also depends on these factors.

In conclusion from the results obtained in our experiment, we recommend the use of *P. canariensis* seedlings for reforestation grown in Arnabat containers with artificial fertilized substrates (fertilization rate 4 g l^{-1} Osmocote).

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