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## Water Relations, Root Growth Potential and Plant Survival of Cold Stored *Pinus radiata* D. Don Seedlings

By

A. MENA-PETITE<sup>1)</sup>, U. ORTEGA-LASUEN<sup>1)</sup>, B. GONZÁLEZ-MORO<sup>1)</sup>, M. LACUESTA<sup>2)</sup>  
& A. MUÑOZ-RUEDA<sup>1)</sup>

**Key words:** Cold storage, *Pinus radiata*, root growth potential, seedling quality, stress, survival, storage temperature, water potential.

### S u m m a r y

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We have investigated the effect of storage conditions (duration: 1, 8 or 15 days; temperature: 4 °C or 10 °C; and root characteristics: bareroot (BR) or soil-plugged root (PR) on the water status, electrolyte leakage and root growth potential (number of new roots and new root length) of *radiata* pine seedlings. We have also examined its effects on plant quality and field performance and their usefulness for detect physiological damage to seedlings affecting plant quality and vitality. Our data show that storage conditions influence the ability of *radiata* pine seedlings to initiate and elongate new roots. The effect was more pronounced in bareroot seedlings than in seedlings with soil around roots, pointing out that the rooting-plug medium provides protection against the desiccation. The water potential at the time of transplanting is a reliable predictor of the ability to generate new roots.

### I n t r o d u c t i o n

To successfully establish coniferous seedlings in the field, it is necessary to understand nursery practice effects on seedling physiology and, hence, field per-

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<sup>1)</sup> Departamento de Biología Vegetal y Ecología, Facultad de Ciencias, Universidad del País Vasco/EHU, Aptdo. 644, E48080 Bilbao, Spain. Fax: 94 4648500. E-mail: [gypmu-rua@lg.ehu.es](mailto:gypmu-rua@lg.ehu.es)

<sup>2)</sup> Departamento de Biología Vegetal y Ecología, Facultad de Farmacia, Universidad del País Vasco/EHU, Aptdo. 450, E-01080 Vitoria, Spain. Fax. 945 130756. E-mail: [gvplacam@vc.ehu.es](mailto:gvplacam@vc.ehu.es)

formance (GROSSNICKLE & BLAKE 1985). In the conifer nurseries of The Basque Country, Northern Spain, seedlings are normally lifted in midwinter (February) and planted immediately or after brief storage. Cold storage of forest planting stock has become a widely used and accepted method of storing plants. However, during lifting, bagged, cold-storage, transport to the planting site and planting, seedlings are subjected to stress conditions (MCKAY 1997). Also, it has been suggested that cold-stored seedlings appear to be more able to avoid transplant shock and early drought (BLAKE 1983). Seedling storage affects the physiological quality of plants which can be reflected in root growth potential, stress resistance and frost hardiness after post-planting (MCKAY 1997).

The purposes of this work were 1) to evaluate the effect of various nursery treatments on the physiological characteristics of planting stock during and after cold storage in the nursery; 2) to ascertain the relationship between root conditions and storage temperature and the combined effects of both parameters on the water relations of radiata pine seedlings, and 3) to analyse the usefulness of these parameters to predict the condition of stored plants and its capacity for short-term post-planting survival.

## Materials and Methods

### Seedling lifting and analyses during cold storage

Plants were raised in Oihanberri nursery (The Basque Country). When plants were nine months old, they were placed in polyethylene bags and immediately carried to the laboratory. Plants were divided into two groups. In one group the roots were surrounded with soil (soil-plugged root seedlings, PR), whereas in the other the roots were free of soil (bare-rooted seedlings, BR). Both groups were packed in polyethylene storage bags and stored in controlled temperature and humidity chambers, during 1, 8 or 15 days, at +4 °C or +10 °C and relative humidity of 80 %.

Predawn xylem water potential was measured as SCHOLANDER & al. 1965, electrolyte leakage in needles and roots was determined using the technique described by MCKAY 1993 and Relative Water Content (RWC) using the equation  $RWC = 100 [(FW-DW)/(TW-DW)]$ .

### Post-planting survival and root regeneration

After the cold-storage period, seedlings were planted in PVC containers (GODET 430, Pépinière Robin, France), containing a mixture of peat moss and vermiculite 1:1 (v/v). Seedlings were grown in a growth chamber for 2 months, in controlled conditions.

Root growth capacity is the ability of a transplanted seedling to produce roots (SUTTON 1980). Root regeneration was determined by measuring the number and root length of new white roots 28 days after transplanting (GROSSNICKLE & BLAKE 1985).

Abbreviations: BR, bare-rooted seedlings;  $\Psi_w$ , water potential; PR, soil-plugged root seedlings; RH, relative humidity; RWC, relative water content; FW: fresh weight; TW: turgid weight; DW: dry weight; NRLR, needle relative leakage ratio; RRLR, root relative leakage ratio; SPP, post-planting survival.

## Results and Discussion

Storage conditions influenced seedling water potential, relative water content (Table 1), and needle and root membrane integrity (Fig. 1). Our data show that

as storage period extends, the water potential of pine seedlings becomes more negative, changing from -0.50 MPa at lifting time to -2.48 MPa after 15 days cold-storage (Table 1), also observed by BALNEAVES & MENZIES 1990, and in the same way, the relative water content also decreases (33% in BR plants stored at 10 °C (Table 1).

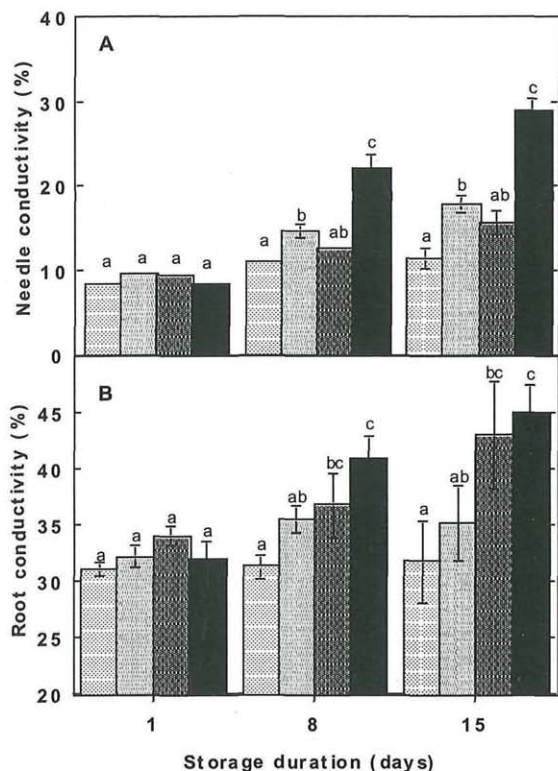


Fig. 1. Effect of storage conditions on the electrolyte leakage of needles (A) or roots (B) in radiata pine seedlings. Soil plugged-root seedlings stored at 4 °C (▨) or 10 °C (▩), and bare-rooted seedlings stored at 4 °C (▧) or 10 °C (■). Each bar represent the means (SE) of three independent experiments, each assayed twice. For a given day, mean values not sharing common letters are significantly ( $p < 0.05$ ).

When we compared means of  $\Psi_w$  and RWC pooled through all treatments, leaf water potential decreased progressively with RWC decline (positive linear relationship ( $r = 0.94$ ), data not shown). The analysis of the electrolyte leakage is a measure of the deterioration of the plant during storage (MCKAY 1993) and its possible impact on field survival. In this regard, we have observed an increase in the electrolyte leakage both in needle (Fig. 1A) and root (Fig. 1B) as cold storage period extends (Fig. 1) (MCKAY & MASON 1991). Though a close relationship between root- and shoot-conductivity was observed (data not shown), the effect of cold storage on shoot was larger than on root.



Table 1. Effect of storage conditions on water potential ( $\psi_w$ ), relative water content (RWC), needle relative leakage ratio (NRLR), root relative leakage ratio (RRLR); root growth potential (after 4 weeks, RGP) and post-planting greenhouse growth survival (after 8 weeks, SPP,) of radiata pine seedlings. C, control plants before storage; 4PR, soil-plugged root seedlings stored at 4 °C for 15 days; 4BR, bare-rooted seedlings stored at 4 °C for 15 days; 10PR, soil-plugged root seedlings stored at 10 °C for 15 days; 10BR, bare-rooted seedlings stored at 10 °C for 15 days.

Treatment	$\psi_w$ (MPa)	RWC (%)	NRLR (%)	RRLR (%)	RGP	SPP
C	-0.53	87.6	7.78	30.6	-	100
4PR	-1.30	84.6	11.34	31.2	15	80
4BR	-1.97	72.4	15.64	43.0	10	40
10PR	-1.45	81.8	17.83	35.1	11	80
10BR	-2.48	58.3	28.80	44.9	3	40

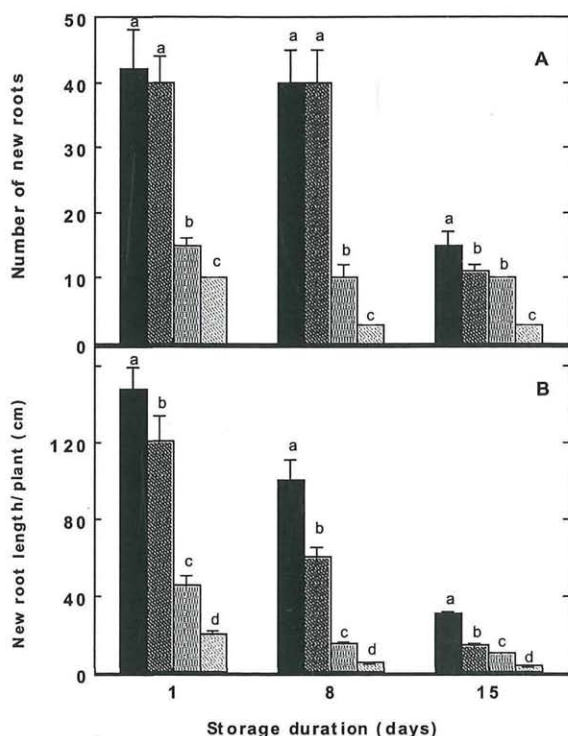


Fig. 2. Effect of storage conditions on root initiation (RGP) (A) or root elongation (B) in radiata pine seedlings. Soil plugged-root seedlings stored at 4 °C (■) or 10 °C (■) and bare-rooted seedlings stored at 4 °C (▨) or 10 °C (▤). Statistical treatment as in Fig. 1.

Root growth potential has often been used as a performance attribute to quality evaluation of conifer seedlings after several storage periods (MATTSSON 1991). Our data show that storage conditions influence the ability of radiata pine seedlings to initiate (Fig. 2A) and elongate (Fig. 2B) roots when placed into fa-

favourable environment for root growth (RITCHIE 1985). After 15 days of storage, the number of new roots was drastically reduced (from about 75 % to 93 %) regardless of temperature and root conditions, although PR seedlings had always more new roots than BR seedlings (Fig. 2). At post-storage  $\Psi_w$  below the value of -1.5 MPa, a 90 % reduction of root extension is produced in *Pinus radiata* seedlings (data not shown), (GIRARD & al. 1997).

A number of post-storage physiological parameters have been used as reliable indicators of seedling field performance after planting. Figure 3 describes the growth-chamber survival of radiata pine seedlings two months after transplanting. We have observed a close relationship between survival capacity and parameters such as length of storage (Fig. 3), post-storage water potential (data not shown), electrolyte conductivity (data not shown; MCKAY & MASON 1991, MCKAY & WHITE 1997), and number and length (data not shown) of new roots. The highly significant ( $r = 0.92$ ) correlation found between root leakage and survival indicates that it principally involves root processes (data not shown).

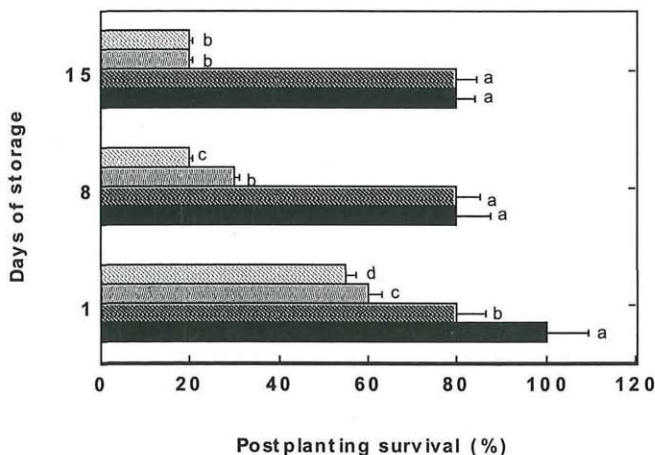


Fig. 3. Effect of storage conditions on postplanting survival of radiata pine seedlings stored for 1 to 15 days. Soil plugged-root seedlings stored at 4°C (■) or 10°C (▨), and bareroot seedlings stored at 4°C (▤) or 10°C (▩). Statistical treatment as in Fig. 1.

The data reported here give us a clear indication of the effect that storage temperature, root conditions and duration of storage have on preplanting quality of radiata pine seedlings. In conclusion, *Pinus radiata* seedlings lifted in middle winter can be cold-stored during periods no longer than two weeks. Storage temperature above 4°C may be detrimental for stock quality, whilst root conditions have a remarkable influence on physiological characteristics. Thus, water potential and root/shoot electrolyte leakage may be good indices of post-storage quality and of initial root and survival potentials in radiata pine seedlings.

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Autor(en)/Author(s): Mena-Petite A., Ortega-Lasuen U., Gonzalez-Moro M. B., Lacuesta M., Munoz-Rueda A.

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