Osmotic adjustment in three-year-old seedlings of five provenances of maritime pine (Pinus pinaster) in response to drought

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Summary Three-year-old seedlings of five provenances of Pinus pinaster Ait. that differed in climatic conditions at their geographical origin were subjected to decreasing soil water availability. The degree of needle osmotic adjustment (OA) was estimated based on logarithmic plots of needle relative water content (RWC) against needle osmotic potential (Ψπ); i.e., \( \ln \text{RWC} \) versus \( -\ln(\Psiπ) \). There were significant differences among provenances in active OA (0.13 to 0.30 MPa for a decrease in RWC to 80%), and a clear negative relationship was found between OA and precipitation (650 to 1280 mm of mean annual rainfall) at the geographical origins of the provenances. A high osmoregulatory capacity contributes to the maintenance of positive turgor at low water potentials. We conclude that OA is one of the mechanisms underlying adaptation to drought in P. pinaster. Solute accumulation was about 2.3 times higher in the provenance from the driest site than in the provenance from the wettest site. The contribution of osmotic adjustment to differences in drought tolerance mechanisms among provenances is discussed.

Keywords: relative water content, turgor maintenance.

Introduction

Osmotic adjustment (OA), defined as the lowering of osmotic potential as a result of net solute accumulation, contributes to drought resistance in many annual and woody plants (Osonubi and Davies 1978, Turner and Jones 1980, Morgan 1984, Colombio 1987, Vivin et al. 1996). Reduced osmotic potential allows the plant to maintain turgor and therefore to maintain cell expansion and high stomatal conductance at low water potentials. Osmotic adjustment is often categorized as a dehydration tolerance adaptation, but it can be considered also as a dehydration avoidance mechanism because it enhances root growth and soil water extraction (Turner 1986, Ludlow and Muchow 1990). Genetic variability in OA capacity has been reported in many crop species (Zhang et al. 1999), suggesting its use as a criterion for selecting drought-resistant plants.

A variety of protocols have been employed to determine the OA capacity of plants subjected to water deficits, hampering comparison of OA data among published studies. The three most commonly used methods are: (1) regression of leaf relative water content (RWC) against leaf osmotic potential (Ψπ) (Morgan 1980); (2) extrapolation of the osmotic potential of water-stressed plants to the rehydrated state (Ludlow et al. 1983); and (3) measurement of osmotic potential of water-stressed plants that have been rehydrated (Blum and Sullivan 1986). Although it is time-consuming to measure RWC and Ψπ at intervals during a drying cycle, Morgan’s method (Morgan 1980) reveals tissue behavior during drought development by partitioning between the concentration effects on osmotic potential resulting from water loss and active solute accumulation. The other methods are more appropriate for screening large numbers of genotypes; however, the measurements must be performed at an equivalent tissue water status, because the magnitude of OA depends on the degree of water stress (Jones and Rawson 1979, Collet and Guehl 1997). Babu et al. (1999) compared these three methods on the same plant material (12 rice cultivars) and found a wider range of OA values estimated by Morgan’s method than by the other methods. Morgan’s method has been used mainly with crop plants (Morgan 1995, Lilley and Ludlow 1996, Lemcoff et al. 1998, Zhang et al. 1999), although Nguyen-Queyrens et al. (2002) recently evaluated the method with Pinus pinaster Ait. cuttings.

Pinus pinaster is widely established in monospecific stands in southwestern Europe and northern Africa. Because of its high stomatal sensitivity to soil water conditions, maritime pine is considered a drought-avoiding species (Granier and Loustau 1994, Picon et al. 1996). In the context of global climate change, breeders are particularly interested in selecting genotypes that can maintain growth performance under drought conditions. Earlier studies of maritime pine revealed variability in ecophysiological response to drought among geographical races (Guyon and Kremer 1982, Nguyen and Lamant 1989, Guehl et al. 1994, Fernandez et al. 1999). Little is known, however, about the adaptive processes involved that could serve as criteria for selection and breeding of more drought-tolerant genotypes.

(1999) reported a significant decrease in osmotic potential at full turgor in *P. pinaster* shoots subjected to water stress. Picon-Cochard and Guehl (1999) did not observe soluble carbohydrate accumulation in *P. pinaster* seedling needles subjected to a soil drying cycle, but they did not estimate OA. Nguyen-Queyrens et al. (2002) recently used Morgan’s method to evaluate OA of *P. pinaster* cuttings and found that the cuttings had a higher capacity for OA compared with crop plants. However, they cautioned that their measurements were probably affected by the slow imposition of water deficit and by the effects of plant height. Furthermore, because the effects of drought on the growth characteristics of the studied genotypes were unknown, the putative benefits of OA for drought adaptation in this species could not be evaluated.

We used Morgan’s method to assess needle OA variation in five ecologically different provenances of maritime pine and to validate the method for use with a coniferous species. Needle turgor response was also analyzed in three of the five provenances during the water potential and osmotic potential measurements. Among genotypes, the ability to actively accumulate solutes and to maintain positive turgor at low water potentials was inversely related to the amount of precipitation at the site of seed origin.

**Materials and methods**

**Plant material and drought treatment**

Five provenances of maritime pine, Landes (France), Porto Vecchio (France), Tamjout (Morocco), Leiria (Portugal) and Tabarka (Tunisia) were chosen based on differences in climatic characteristics at the sites of seed origin (Table 1). Seeds were provided by the Forest Genetics and Breeding Laboratory (INRA, Bordeaux-Cestas, France), and were sown in a greenhouse during spring 1996. After several months, the seedlings were transplanted to 4-l containers filled with sand:peat:cork (1:1:1 v/v) and placed in a nursery. At the beginning of 1999, 9 to 11 trees per population were transplanted to 4-l pots in the experimental trees every second day.

Needle predawn water potential (Ψ$_{wp}$) was measured with a Scholander pressure chamber (Scholander et al. 1965, Ritchie and Hinckley 1975) five times during the treatment period ($t_0$, $t_1$, $t_3$, $t_5$, $t_6$, $t_8$ and $t_{10}$) in three provenances (Landes, Porto Vecchio and Tamjout). At each measurement time, two 1-year-old needles per tree of 9–11 trees per provenance were taken from the main shoot and measured to give a tree mean value of Ψ$_{wp}$.

At the same time, but for all the provenances, 12 1-year-old needles per tree (six pairs of needles per tree) were harvested from the main shoot. Within a subsample, three pairs were divided to give three needles for osmotic potential (Ψ$_o$) measurement and three needles for relative water content (RWC) measurement. The mean value from the two subsamples per tree was used to characterize each tree. For Ψ$_o$ measurement, the needles were cut into small pieces (2–3 mm long) and...
placed in a 2-ml syringe, and needle sap was squeezed out after freezing (in liquid nitrogen, −196 °C) and thawing. At the end of the drought treatment, the dryness of the needles of several samples prevented extraction of a sufficient quantity of sap. Sap osmolality was measured in 10-µl sap samples with a vapor pressure osmometer (Vapro R Model 5520, Wescor, Logan, UT). The RWC was obtained by weighing the needles just after harvest (FW = fresh mass) and then placing them in water at 5 °C in darkness for 24 h to determine fresh mass at full hydration (FW100). The proximal end was previously submerged and severed to avoid formation of air bubbles in the vessels that could prevent complete rehydration. Finally, needle dry mass (DW) was determined (70 °C, 48 h) and RWC was calculated as RWC = (FW − DW)/(FW100 − DW).

Active osmotic adjustment (OA) was determined from bi-phasic plots of ln RWC against −ln(−Ψπ) (Morgan 1980, 1992, Lilley and Ludlow 1996, Lilley et al. 1996, Wright et al. 1997, Lemcoff et al. 1998) as the trees were subjected to gradual water stress. The first phase corresponds to a change in osmotic potential (Ψπ) at constant RWC (no water loss); this solute accumulation is called “absolute OA.” During the second (falling) phase, water loss occurs: an increase in solute concentration exceeding that resulting from a decrease in water content (the passive component) is evidence of active osmotic adjustment. In the case of a passive osmometer, ln RWC = −A ln(−Ψπ) + B, where A = 1 (Morgan 1980). When there is active OA, A is less than 1. The degree of osmotic adjustment for each population was therefore determined from the relationship between ln RWC and −ln(−Ψπ) as the trees were subjected to gradual water stress (Morgan 1995).

The amount of solute accumulated at 80% RWC (OA80) was calculated as the difference between initial osmotic potential Ψπi (measured at t0) and osmotic potential at 80% RWC (Ψπ80, derived from the function fitted to the data). Each variable was corrected for the concentration effect of dehydration by multiplying by RWC, (initial relative water content) and 0.8:

$$OA_{80} = RWC_i - 0.8Ψ_{π80}$$

A relative water content of 80% was chosen because it was near the threshold for loss of turgor.

Statistical analysis

Analyses of variance were performed with SAS software (1987; SAS Institute, Cary, NC) to determine the effect of provenance on measured or calculated parameters. All statistical comparisons were considered significantly different at P < 0.05 and means were compared by the Duncan test. Relationships between variables were analyzed by simple linear regression using SAS software, or by two linear regression using SigmaPlot software procedures (1999; SPSS, Richmond, CA). Slopes of linear regressions were compared in pairs based on the significance of the interaction between provenance and the x-axis variable.

Results

Mean seedling height by provenance is given in Table 1. Provenances Porto Vecchio and Leiria had significantly taller seedlings than the other provenances.

Time courses of needle predawn water potential (Ψπwp) and relative water content (RWC)

Predawn RWC values at t0 were low (90–93%), indicating slight stress. Measurements were made about 24 h after water had been supplied and the plants had been subjected to a sunny day in the greenhouse.

Needle Ψπwp values are presented in Table 2. From a mean value among provenances of −0.45 MPa at t0, Ψπwp dropped to final values of −1.8, −2.0 and −2.8 MPa for Porto Vecchio, Landes and Tamjout provenances, respectively. Among provenances, Tamjout exhibited the most negative Ψπwp values after 3 weeks of water deficit, whereas Ψπwp of the two French provenances showed the same pattern over time. Tabarka was also among the provenances showing the greatest decrease in RWC over the drying cycle (Table 2), which could explain the yellowish and rather dry needles observed on trees of the Tamjout provenance at the end of the experiment. Among provenances, Tabarka for the whole treatment and Leiria from t0 maintained the highest needle RWC values.

Tree height had no effect on Ψπwp; however, a negative relationship was found with RWC, but only under conditions of slight (RWCi, week t0) or mild stress (week t3). The tallest trees belonging to the Porto Vecchio and Leiria provenances had low needle relative water contents.

Table 2. Time course of predawn needle water potential (Ψπwp, mean ± SD, n = 9 to 11) for the provenances Landes, Porto Vecchio and Tamjout, and of relative water content (RWC) for all provenances. For each week, means with the same letter are not significantly different at P = 0.05.

<table>
<thead>
<tr>
<th>Provenance</th>
<th>Ψπwp (MPa)</th>
<th>RWC (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>t0</td>
<td>t3</td>
</tr>
<tr>
<td>Landes</td>
<td>−0.46 a</td>
<td>−0.55 a</td>
</tr>
<tr>
<td>Porto Vecchio</td>
<td>−0.39 a</td>
<td>−0.55 a</td>
</tr>
<tr>
<td>Tamjout</td>
<td>−0.46 a</td>
<td>−0.64 a</td>
</tr>
<tr>
<td>Tabarka</td>
<td>93.3 a</td>
<td>91.7 ab</td>
</tr>
</tbody>
</table>
**Water relations**

The ln RWC data for each provenance are plotted against the $-\ln(-\Psi_{\pi})$ data in Figure 1. During the first phase of "absolute osmoregulation," $\Psi_{\pi}$ fell from about $-1.4$ MPa to $-1.7$ MPa, whereas RWC showed no decline. In the second (falling) phase, both $\Psi_{\pi}$ and RWC declined. A broken-stick regression was fitted to the whole data set for each provenance (Figures 1 and 2), while holding the slope of the first phase to zero. The regression coefficients (Table 3) were low for the Leiria and Tabarka provenances (0.6 and 0.7, respectively). We note, however, that few data were available for these provenances for the second phase (Figure 1), probably because the applied water deficit was too weak to cause a substantial decrease in RWC in these two provenances (Table 2).

In the first phase, the calculated constant value of RWC was significantly lower for the Porto Vecchio and Leiria provenances (89.5%) than for the other provenances (91.5%), in accordance with the measured RWC$_{i}$ values (Table 3). The initial osmotic potential ($\Psi_{\pi_{i}}$) varied between provenances, from $-1.59$ MPa in Tabarka and Tamjout to $-1.46$ MPa in Landes. The break point (i.e., the $\Psi_{\pi}$ value at the intersection of the two linear phases) ranged from $-1.73$ MPa (Tabarka) to $-1.57$ MPa (Landes). Solute accumulation during the first phase of absolute OA was estimated from the difference between the break point and $\Psi_{\pi_{i}}$ (the “length” in Table 3), leading to a narrow range of values from 0.09 MPa (Tamjout) to 0.15 MPa (Leiria).

During the second phase, marked departure from a passive osmometer (slope value less than 1) occurred for all populations, indicating solute accumulation as a result of osmotic adjustment. Provenances differed in OA capacity (Table 3): the highest OA capacity was found in the Tamjout provenance with a slope of 0.49, whereas the lowest OA capacity was observed in Landes, with a slope of 0.85. The other provenances showed intermediate behavior. The slope decreased with decreasing mean annual rainfall at the site of seed origin (Figure 3): the drier the site, the smaller the calculated slope, and thus the greater the osmotic adjustment capacity of the needle tissue.

The turgor response to drought depended on the relationship between needle water potential ($\Psi_{wp}$) and needle osmotic potential ($\Psi_{\pi}$, Figure 4). Fitted linear regressions were traced and the slopes differed between the three studied provenances (Table 4). The difference between the response line and the 1:1 line provides an estimate of the turgor potential. Turgor loss, where $\Psi_{\pi} \approx \Psi_{wp}$, occurred at water potential values ($\Psi_{0}$) of $-1.80$, $-2.00$ and $-2.20$ MPa for the Landes, Porto Vecchio and Tamjout provenances, respectively (Table 4). The Tamjout provenance was thus able to maintain positive needle turgor at a lower water potential than the Landes or Porto Vecchio provenances. The ability to maintain turgor ability was positively associated with OA capacity.

Relative water contents derived from the function fitted to the data (Table 3), and evaluated at $\Psi_{0}$, were 81.2, 79.2 and 80.1.
80% for the Landes, Porto Vecchio and Tamjout provenances, respectively. To compare solute accumulation (OA80) between provenances, a relative water content of 80% was chosen because it was near the region beyond which turgor was lost. Among provenances, OA80 (Table 3) showed the same ranking as OA capacity. Solute accumulation in needle tissue was about 250% greater in Tamjout (0.30 MPa), and about 150% greater in Tabarka, Leiria and Porto Vecchio (0.18 to 0.23 MPa) than in Landes (0.13 MPa).

**Discussion**

The In RWC/–ln(Ψπ) relationship for *Pinus pinaster* was biphasic. Such a relationship is not universal. For some genotypes of wheat, peas, sunflower and rice (Morgan 1980, Rodriguez-Maribona et al. 1992, Chimenti and Hall 1993, Lilley and Ludlow 1996), the first phase (absolute osmotic adjustment with no water loss) has not been observed. The presence of the first phase in *P. pinaster* indicates that osmotic adjustment begins early in the stress period. Nevertheless, the first phase of osmotic adjustment in *P. pinaster* was short and the range of variation narrow (0.09 to 0.15 MPa) compared with values reported by Lilley and Ludlow (1996) for 61 rice lines (0.2 to 1.5 MPa), by Lemcoff et al. (1998) for 14 maize hybrids (0.4 to 0.5 MPa) and by Chimenti and Hall (1993) for 40 sunflower cultivars (0.2 to 0.9 MPa).

The observed break points in the ln RWC/–ln(Ψπ) relationships of the *P. pinaster* provenances were more negative than in maize (Chimenti and Hall 1993) or sunflower (Lemcoff et al. 1998) (−1.23 to −0.9 MPa and −1.5 to −0.7 MPa, respectively), but within the range observed for rice lines (−2.84 to −1.52 MPa) (Lilley and Ludlow 1996). The low break points in *P. pinaster* should confer delayed water loss in response to drought; however, no correlation was found with rainfall characteristics of the sites of seed origin.

Active osmotic adjustment occurred in all five provenances during tissue water loss and significant differences between provenances were observed, even though the range in slope of the second phase (0.49 to 0.85% MPa–1) was narrower than in rice lines (0.58 to 1.16% MPa–1), maize hybrids (0.16 to 0.92% MPa–1) or sunflower cultivars (0.5 to 1.6% MPa–1). A clear positive relationship was found between OA and precipitation at the site of seed origin, suggesting that osmotic adjustment has a role in drought adaptation in *P. pinaster*. Rainfall, however, may not always be the only relevant descriptor of climate. Several authors have demonstrated that the rate of drought development also influences the expression of OA (Jones and Rawson 1979, Collet and Guehl 1997, Babu et al. 1999, Shangguan et al. 1999). In 3-year-old seedlings of *Quercus petraea* L. ex Liebl. (Collet and Guehl 1997), the capacity to adjust osmotically was detected in the water deficit treatment of −0.013 MPa day−1, but not in the −0.05 MPa day−1 treatment. In our study, the rate of drought development calculated roughly as linear in time over the treatment period varied from −0.023 MPa day−1 (Porto Vecchio) to −0.033 MPa day−1 (Tamjout). Although trees of the Tamjout provenance begin to adjust osmotically in response to drought, the range of variation is narrow (0.09 to 0.15 MPa) compared with values reported by Lilley and Ludlow (1996) for 61 rice lines (0.2 to 1.5 MPa), by Lemcoff et al. (1998) for 14 maize hybrids (0.4 to 0.5 MPa) and by Chimenti and Hall (1993) for 40 sunflower cultivars (0.2 to 0.9 MPa).

**Table 3.** Initial values (week *t*0) of osmotic potential (Ψπi) and relative water content (RWCi), and linear regression parameters of ln RWC against –ln(Ψπ). Within columns, parameters with the same letter are not significantly different at *P* = 0.05.

<table>
<thead>
<tr>
<th>Provenance</th>
<th>Week <em>t</em>0 (n = 9 to 11)</th>
<th>Broken-stick regression analysis of ln RWC against –ln(Ψπ) (n = 34 to 44)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ψπi (MPa)</td>
<td>RWCi (%)</td>
</tr>
<tr>
<td>Landes</td>
<td>−1.46 a</td>
<td>91.8 ab</td>
</tr>
<tr>
<td>Tabarka</td>
<td>−1.59 b</td>
<td>93.3 a</td>
</tr>
<tr>
<td>Leiria</td>
<td>−1.52 b</td>
<td>90.2 b</td>
</tr>
<tr>
<td>Porto Vecchio</td>
<td>−1.49 a</td>
<td>90.1 b</td>
</tr>
<tr>
<td>Tamjout</td>
<td>−1.59 ab</td>
<td>91.8 ab</td>
</tr>
</tbody>
</table>
were subjected to the most severe water stress, the Tamjout population expressed the highest OA among provenances, indicating that the rate of drought development did not influence the occurrence of OA.

Calculation of OA80 has the advantage of combining two components of osmotic adjustment—the break point and the slope of the second phase—to compare provenances subjected to a comparable degree of stress. Such a parameter has been reported only for herbaceous species, and calculated at 70% RWC (OA70), a water status value around turgor loss in these species. This lower reference RWC value probably contributed partly to the large variation in OA observed among cultivars or lines of crop plants: 0.7 to 1.3 MPa in pigeon pea (Flower and Ludlow 1987), 0.8 to 1.7 MPa in sorghum (Basnayake et al. 1993), 0.4 to 1.5 MPa in rice (Lilley and Ludlow 1996), and 0.14 to 1.35 MPa in wheat (Blum et al. 1999). Based on the pressure–volume curve method on 1-year-old Pinus pinaster shoot seedlings, Fernandez et al. (1999) found a maximum difference of 0.4 MPa between osmotic potentials at full turgor (\(\Delta \Psi_{w100}\)) before and after water stress treatment. Although Fernandez et al. (1999) did not report RWC values at harvest, this difference is in agreement with our OA80 data (0.13–0.30 MPa). For comparison, other values of \(\Delta \Psi_{w100}\) between controlled and water-stressed woody seedlings are: 0.2 MPa for P. mariana (Tan and Blake 1997), 0.6 MPa for Quercus petraea and Quercus robur L. (Collet and Guehl 1997, Thomas and Gausling 2000), 0.15 MPa for Pinus halepensis Mill. (Tognetti et al. 1997), and 0.2 MPa for Eucalyptus Globulus Labill. (Pita and Pardos 2001).

In our study, differences in OA capacity were consistent with differences in the water potential at which leaves reached zero turgor (\(\Psi_0\)). Based on Morgan’s method, similar conclusions have been reported for wheat (Morgan 1980) and Brassica (Wright et al. 1997). Our result therefore confirms that active solute accumulation maintains turgor during a decline in soil water availability. Morgan’s method for OA determination also appeared to be validated for coniferous species. Water potentials calculated at turgor loss (\(\Psi_0\)) were close to values previously measured in P. pinaster seedlings by the pressure–volume curve method, either using needles (\(\Psi_0 = -2\) MPa for diverse provenances, and \(\Psi_0 = -2.5\) MPa for Tamjout; Sarrauste 1982), or using entire shoots (\(\Psi_0 = -2.47\) to \(-1.73\) MPa for several provenances, Fernandez et al. 1999). Furthermore, similar RWC values at turgor loss were found by Fernandez et al. (1999) (81.3–84.9%) and in our study (79.2–81.2%).

Despite a high needle OA capacity, the Tamjout provenance lost needle water more rapidly than the other provenances, as indicated by the decrease in RWC. We note, however, that the experiment was conducted in containers, and thus in a limited soil volume. Earlier studies (Sarrauste 1982, Guehl et al. 1994) demonstrated higher root biomass proportions for Tamjout, which accelerated soil drying. Furthermore, Nguyen and Lamant (1989) found that OA in root tips of young seedlings cultivated in polyethylene glycol solutions was higher for Tamjout than for Landes. Osmotic adjustment in roots is be-

Table 4. Linear regression analysis of needle osmotic potential (\(\Psi_z\)) against needle predawn water potential (\(\Psi_{wp}\)) for the Landes, Porto Vecchio and Tamjout populations. The intercept of the regression line with the 1:1 line is the water potential at turgor loss (\(\Psi_0\)).

<table>
<thead>
<tr>
<th>Provenance</th>
<th>n</th>
<th>Regression line</th>
<th>Comparison of slope ((P = 0.05))</th>
<th>(\Psi_0) (MPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Landes</td>
<td>37</td>
<td>(\Psi_z = 0.26\Psi_{wp} - 1.33 (r^2 = 0.64))</td>
<td>a</td>
<td>-1.80</td>
</tr>
<tr>
<td>Porto Vecchio</td>
<td>36</td>
<td>(\Psi_z = 0.32\Psi_{wp} - 1.37 (r^2 = 0.81))</td>
<td>ab</td>
<td>-2.01</td>
</tr>
<tr>
<td>Tamjout</td>
<td>37</td>
<td>(\Psi_z = 0.36\Psi_{wp} - 1.41 (r^2 = 0.82))</td>
<td>b</td>
<td>-2.20</td>
</tr>
</tbody>
</table>
lieved to facilitate soil water extraction by increasing the water potential gradient between root tissue and soil, and by maintain-
ing root growth and deeper soil penetration in drought con-

High needle OA capacity should delay stomatal closure during water deficit, as already observed in many woody species (Tan and Blake 1997, White et al. 2000, Pita and Pardos 2001). In P. pinaster, this is supported by some results on seedlings and on adult trees (Guyon and Kremer 1982, Guehl et al. 1994, André 1999) showing the tendency of provenances from drier sites to maintain stomatal activity under conditions of soil or atmospheric water deficit. In addition, J.M. Guehl and M.B. Triboulot (UMR Ecologie et Ecophysiology Forestière, INRA, F-54280, Champenoux, France, personal communication) recently found significantly higher hydraulic conductance and leaf specific hydraulic conductance in Tamjout compared with Landes: these traits are known to contribute to the maintenance of stomatal aperture without affecting leaf water status.

For the Tamjout provenance, high OA in both leaves and roots should promote soil water extraction and whole-plant water circulation, and so help minimize plant dehydration, as long as there is soil water to extract. When the water reserve is consumed, or in the case of severe water deficit, other mecha-
nisms, especially stomatal closure, presumably come into op-
eration. A similar strategy has been observed by Grieu et al. (1988) who reported that Pseudotsuga macrocarpa (Vasey) Mayr. develops a deep root system with high solute concentra-
tions, and presents a less sensitive stomatal response to drought than Pseudotsuga menziesii (Mirb.) Franco. Different drought avoidance strategies were also found among Larix decidua (Mill.), Picea abies (L.) Karst and Pinus cembra L. (Badalotti et al. 2000), with L. decidua developing osmotic ad-
justment in shoots and maintaining high daily sap flux during summer.

In conclusion, we found intraspecific variation in needle os-
motic adjustment of P. pinaster as measured by Morgan’s method. This variation was related to the dryness of the site of seed origin, indicating an adaptive role of needle OA to drought. Geographical populations probably developed di-
verse strategies, some mainly limiting water loss by stomatal closure (Landes), others mainly favoring water circulation with the help of an integrated whole-plant strategy, of which osmotic adjustment represents one mechanism (Tamjout). There are indications that the growth of trees of provenances from drier habitats is less affected by low water availability (Sweet and Thulin 1962, Hopkins 1971, Destrem et al. 1974, Guyon and Kremer 1982). Overall, OA may be a useful criterion for selecting P. pinaster genotypes for satisfactory growth under drought conditions. However, because of ini-
tially low productivities, these provenances could prove less vigorous in the long term. Thus, breeders should consider both adaptive and growth traits, depending on environmental con-
text and practical objectives.

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