

Original article

Influence of soil drying on leaf water potential, photosynthesis, stomatal conductance and growth in two black pine varieties

François Lebourgeois^{a*}, Gérard Lévy^b, Gilbert Aussenac^c,
Bernard Clerc^c, François Willm^c

^aÉquipe phytoécologie, Unité d'écophysiologie forestière, Inra Nancy,
54280 Champenoux, France

^bÉquipe sol et nutrition, Unité d'écophysiologie forestière, Inra Nancy,
54280 Champenoux, France

^cÉquipe bioclimatologie, Unité d'écophysiologie forestière, Inra Nancy,
54280 Champenoux, France

(Received 12 June 1997; accepted 20 October 1997)

Abstract – The aim of this study was to examine the influence of long-term soil water deficit on growth and physiological processes of two black pine varieties (*Pinus nigra* ssp. *laricio* var. *Corsicana* and *Pinus nigra* ssp. *laricio* var. *Calabrica*). Three-year-old seedlings grown in large boxes (volume: 1.62 m³) were subjected to a prolonged summer drought (99 days from the end of June until the end of September) and photosynthesis (*A*), stomatal conductance (*g_w*), water status and growth were measured. No marked differences arose between Corsican and Calabrian pines feature to drought. At least in their juvenile stage, both varieties exhibited a 'drought-avoidance strategy' characterized by an efficient stomatal control of transpirational water loss. This result is consistent with previous studies on *Pinus nigra* and confirm the water stress adaptation of this collective Mediterranean species. Because a significant decrease of *g_w* (about 30 %) was observed with no obvious variation in Ψ_{wp} , the data suggested that predawn water potential was not the best indicator to precociously detect water stress. However, both *A* and *g_w* reduced to nearly zero as soon as the threshold value of $\Psi_{wp} = -1.6$ MPa was reached (respective values 0.5 $\mu\text{mol m}^{-2} \text{s}^{-1}$ and 11 $\text{mmol m}^{-2} \text{s}^{-1}$). Because most fine roots were within the upper 40 cm of the soil, a superficial soil desiccation has probably induced rapid stomatal closure, triggered by a biophysical and/or biochemical signal from the desiccated roots to the leaves. Embolism seems not to be responsible for the effect of drought on physiological processes, because the minimum value of Ψ_{wp} observed at the end of the drying cycle (-2.5 MPa) remained higher than the threshold inducing a significant xylem cavitation for these varieties (-4 MPa). Summer drought significantly reduced annual stem diameter (-20 %) and needle length (-25 %), but not stem elongation. Total elaborated dry weight was reduced about 45 %. Seedlings grown in the dry regime

* Correspondence and reprints
E-mail: lebourgeois@nancy.inra.fr

reduced belowground growth proportionally more than aboveground growth, causing a significant decrease in the R/S ratio. Such a result, which diverges with classical models of whole plant biomass partitioning, might be partially explained by seasonal pattern in the root growth which typically has its most important peak in mid-summer, period of maximum drought in our study. With the parameters studied here, the expression of the genetic characteristics between varieties in drought tolerance appeared to be limited. Thus, further investigations could be undertaken to learn about drought feature at cell and molecular levels. (© Inra/Elsevier, Paris.)

Corsican pine / Calabrian pine / leaf conductance / photosynthesis / drought / root/shoot / growth / biomass partitioning / *Pinus nigra*

Résumé – Influence de la sécheresse sur le potentiel hydrique foliaire, la photosynthèse, la conductance stomatique et la croissance de deux variétés de pins noirs. L'objectif de cette étude était d'analyser l'influence d'une sécheresse édaphique prolongée sur la croissance et le comportement écophysio-logique de deux variétés de pins noirs (*Pinus nigra* ssp. *laricio* var. *Corsicana* et *Pinus nigra* ssp. *laricio* var. *Calabrica*). Des plants de 3 ans, cultivés dans des grandes cuves (volume : 1,62 m³), ont été soumis à une sécheresse de 99 j (de fin juin à fin septembre) pendant laquelle la photosynthèse, la conductance stomatique, l'état hydrique et la croissance des plants ont été mesurés. Au moins dans leur stade juvénile, les deux variétés ont présenté la même stratégie « d'évitement » caractérisée par un contrôle stomatique efficace de la transpiration. Ce résultat est conforme à ceux obtenus sur les pins noirs et confirme la stratégie commune d'adaptation à la sécheresse de cette espèce méditerranéenne. La fermeture stomatique rapide, avant que le statut hydrique ne soit affecté, suggère que le potentiel hydrique de base n'est pas le meilleur paramètre pour détecter précocement le stress. Cependant, la photosynthèse et la conductance stomatique se sont stabilisées à des valeurs très faibles dès que le potentiel hydrique de base a atteint le seuil de -1,6 MPa (respectivement 0,5 $\mu\text{mol m}^{-2} \text{s}^{-1}$ et 11 $\text{mmol m}^{-2} \text{s}^{-1}$). Le système racinaire superficiel (densité maximale de racines dans les 40 premiers centimètres) a probablement joué un rôle déterminant en détectant précocement la sécheresse puis en transmettant un signal chimique et/ou biophysique des racines sèches jusqu'aux feuilles. La cavitation ne semble pas avoir joué un rôle majeur dans les comportements observés étant donné que la plus faible valeur mesurée de Ψ_{wp} (-2.5 MPa) est restée supérieure au seuil d'embolie défini pour cette espèce (-4 MPa). L'accroissement radial et la longueur des aiguilles ont été significativement réduits par la sécheresse estivale (respectivement de -20 et de -25 %) alors que l'élongation de la pousse terminale n'a pas été affectée. La réduction totale de matière sèche élaborée a été d'environ 45 %. Les plants soumis à la sécheresse ont alloué moins de ressources à la croissance racinaire qu'à la croissance aérienne induisant une diminution significative du rapport R/S. Un tel résultat diverge des modèles classiques d'allocations de matières, mais peut partiellement s'expliquer par le rythme saisonnier de croissance des racines qui atteint son optimum au milieu de l'été ; période de sécheresse édaphique maximale dans notre étude. Avec les paramètres étudiés ici, l'expression des variations génétiques entre les deux variétés dans le comportement vis-à-vis de la sécheresse est apparue limitée. Ainsi, d'autres travaux devraient être envisagés afin de mieux cerner les régulations aux niveaux cellulaire et moléculaire. (© Inra/Elsevier, Paris.)

pin laricio de Corse / pin laricio de Calabre / conductance stomatique / photosynthèse / sécheresse / R:S / croissance / répartition de biomasse / *Pinus nigra*

1. INTRODUCTION

Calabrian pine (*Pinus nigra* ssp. *laricio* var. *Calabrica*) and Corsican pine (*Pinus nigra* ssp. *laricio* var. *Corsica*) are two varieties of the Mediterranean collective species *Pinus nigra* [10, 50]. In

their natural stands, the ecological growth conditions are fairly similar with a wide range of altitudinal and soil conditions and a hot, dry summer [16, 17, 19]. The morphological differences between these varieties are small, particularly among mature trees, and apply to bark structure, leaf mor-

Table I. Characteristics and nutrient availability of the growing substrate. S/T 100 represents the saturation rate of each horizon. S = sum of exchangeable cations ($\text{cmol}^+\text{kg}^{-1}$); T = exchange capacity of cations ($\text{cmol}^+\text{kg}^{-1}$).

Depth	Texture (%)			Organic matter ($\text{g}\cdot\text{kg}^{-1}$)	C/N	pH	Exchangeable cations ($\text{cmol}^+\text{kg}^{-1}$)			S/T 100	P_2O_5 ($\text{g}\cdot\text{kg}^{-1}$)
	sand	loam	clay				Ca	Mg	K		
0–10 cm	22.5	52	25.5	30.8	11.9	5.1	4.3	0.48	0.25	66.2	0.16
30–50 cm	21	53.6	25.4	28.2	12.7	4.8	2.9	0.35	0.2	49.3	0.17

2.2. Water supply regimes

The experimental plots consisted of two control plots and two dry plots.

Irrigated plots: maintained permanently near field capacity by frequent manual watering. Soil water content was measured weekly in each box at a depth of 40 and 60 cm with two tensiometers.

Droughted plots: to extrapolate to natural conditions, a prolonged summer drought was imposed. Drought began on 22 June 1993 (Julian day 174). The rewatering to field capacity occurred on 2 October (Julian day 276), after 99 days of drought. Seedling recovery was sampled 3, 6 and 10 days after rewatering. Soil water content was measured weekly in each box at a depth of 40 and 60 cm with two psychrometers. Unfortunately, due to technical problems, soil water potential could only be measured from day 36 of the drying cycle.

2.3. Ecophysiological measurements

Ecophysiological measurements were carried out on 16 3-year-old seedlings (four plants of each variety per treatment) representative of the sample. The leaf water potential was measured weekly from 22 June onwards on needles using a pressure chamber [46]. Needles were sampled in the middle of the annual shoot just prior to dawn (predawn leaf water potential, Ψ_{wp}) and at 1 pm solar time when the sun was at its zenith (midday leaf water potential, Ψ_{wm}).

Gas exchange measurements were performed using a portable gas exchange measurement system (LiCor 6200, LiCor, Lincoln, NE, USA) under natural climate. Environ-

mental conditions during the season were: $\text{PPFD} = 1054 \pm 436 \mu\text{mol m}^{-2} \text{s}^{-1}$; $T_{\text{air}} = 25.2 (\pm 2.8) ^\circ\text{C}$; air CO_2 concentration (C_a) = $346.6 (\pm 14.5) \mu\text{mol mol}^{-1}$. Net CO_2 assimilation rate (A , $\mu\text{mol m}^{-2} \text{s}^{-1}$) and stomatal conductance to water vapour (g_w , $\text{mmol m}^{-2} \text{s}^{-1}$) were calculated with the classical equations of Caemmerer and Farquhar [9]. Gas exchange measurements were made at the same time as midday leaf water potential. At the end of the experiment, calculations were performed on the basis of the total projected needle area of the branches using a video camera coupled to an image analyser (ΔT Devices, Cambridge, UK).

2.4. Growth measurements

At the end of the first growing season (October 1992), annual stem elongation and total height were measured.

In 1993, the bud expansion was first observed in early April. The first stem measurement was made in mid-June before the beginning of the moisture stress treatment. After the summer drought, total height, total stem elongation, basal stem diameter and length of new needles were measured. The needle length was measured to the nearest mm from the point of fascicle sheath insertion in the axil of a subtending cataphyll (bract) to the needle top. For each plant, ten needles were randomly chosen in the middle of the current-year shoot.

In order to estimate the biomass distribution in the various organs (needle, stem and root) during an annual vegetative cycle and to quantify the below- and aboveground growth responses to the water stress treatment, two boxes (one control plot and one dry plot) were harvested at the end of the drought. Because

phology and anatomy, as well as cone morphology [3, 21, 22]. Despite the increased interest in their use for afforestation, their ecological plasticity outside the area where they are indigenous is not well known. In France, Corsican pine is generally recommended on more or less acidic soils, whereas Calabrian pine seems to be able to withstand soils with temporary water table [23, 36]. However, both varieties are mixed in most managed crops.

Pinus nigra water stress tolerance is also poorly understood and comparative studies between subspecies and varieties of this species are rather rare. Becker [7], studying transpiration and drought behaviour of 3-year-old seedlings of some coniferous species (Eastern White pine, Douglas fir, Norway spruce and Corsican pine), showed that Corsican pine had the best water use efficiency (shoot biomass increment versus transpired water during the experiment). Aussenac and Granier [5] and Aussenac and Valette [6] showed that in response to soil water depletion, 15-year-old black pine trees (*Pinus nigra* ssp. *nigricans*) rapidly decreased transpiration and photosynthesis, with these processes being totally inhibited at a relatively high predawn needle water potential (-1.6 to -1.7 MPa). For the subspecies 'laricio', Aussenac [4], using excised shoots subjected to a desiccation, described a similar feature with a significant stomatal closure when predawn needle water potential reached values around -1.2 to -1.4 MPa. A dendroecological study carried out on 1 808 mature Corsican pine in western France also showed that drought was one of the major environmental factors influencing and limiting radial growth and wood productivity. The current decline of trees has been mainly related to repeated severe drought events that have occurred since the end-1960s in this region [35].

In order to compare the water stress sensitivity of both varieties and also to

highlight the underlying mechanisms to the growth decrease observed in mature stands, an ecophysiological study was undertaken. In this present work, plant water status, gas-exchange responses and growth of both irrigated and droughted seedlings were measured and analysed.

2. MATERIALS AND METHODS

2.1. Experimental design

The experimental design was set up at the Inra Research Centre of Nancy (Lorraine, northeast France). It consisted of four large partially buried boxes (depth: 100 cm; width: 144 cm; volume: 1.62 m³). Thus, the root development of plants was not limited or at the least less confined than if they had been in small-sized containers. These boxes were filled with 10 cm of gravel at the bottom to improve water drainage, and 80 cm of a sifted sandy clay loam soil from the horizon A1/A2 of a Dystric Cambisol (Food and Agriculture Organization classification) from the Haye Forest (France). The characteristics of the growing substrate are presented in table 1. No fertilization was applied because chemical composition corresponded to optimal conditions of plant nutrition [36].

At the beginning of March 1992, 2-year-old (2 + 0) seedlings (seed origin: Corsica and Calabrica; average height: 10 cm) from the Forest Research Center's nursery were planted in staggered rows (35 plants of each variety per box). In order to avoid any possible inter-variety competition effects, varieties were not mixed in the boxes. During this first growing season (1992), all trees were grown in open conditions, and kept well-watered by natural and manual irrigation. No herbicidal or fertilizing treatment was applied.

At the beginning of June of the following year (1993), a transparent polyethylene tunnel opened at its extremities was installed in order to intercept rainfall and to maintain sufficient ventilation during hot summer days.

of the absence of blocking (box) effects, the two boxes were randomly chosen. For the root system, each plant was manually and carefully uprooted, and to avoid any error due to wall effect, plants near the wall were eliminated. However carefully applied, this method did not allow us to sample all fine roots. Nevertheless, direct and visual observations showed that the root system remained superficial (maximum root density above 40 cm) for both treatments and varieties. Only the largest roots reached the deep soil horizon (1 m). All the samples were also oven-dried at 80 °C for 48 h. The biomass partitioning among the plant compartments was assessed by determining a) the leaf mass ratio (LMR, leaf dry mass/whole plant dry mass, g g^{-1}), b) the stem mass ratio (SMR, stem dry mass/whole plant dry mass, g g^{-1}), c) the root mass ratio (RMR, root dry mass/whole plant dry mass, g g^{-1}), d) the root/shoot ratio (R/S, root dry mass/shoot dry mass, g g^{-1}). The shoot dry weight equalled the dry weights of leaves plus stems plus branches.

One-way and two-way analyses of variance (ANOVA followed by Fisher's PLSD test) were used to evaluate the significance of the single and interactive effects of drought and varieties.

3. RESULTS

3.1. Plant water status

No marked differences arose between Corsican and Calabrian pine leaf water potential response to drought (figure 1). From 15 days onwards, water stress increased gradually with predawn water potentials decreasing around -0.2 MPa per week in both varieties. After 73 days, predawn water potentials reached stable values around -2.5 MPa . The decrease in Ψ_{wp} was closely related to soil water content (figure 2).

In watered treatments, Ψ_{wp} and Ψ_{wm} ranged from -0.22 to -0.55 MPa (average: $-0.32 \pm 0.08 \text{ MPa}$) and from -0.7 to -2.2 MPa (average: $-1.2 \pm 0.4 \text{ MPa}$), respectively. These values corresponded to the common observed data for the *Pinus* species [44].

After rewatering (R), seedlings recovered rapidly. Three days after rewatering, water potentials were again equivalent to those of irrigated plants (figure 1).

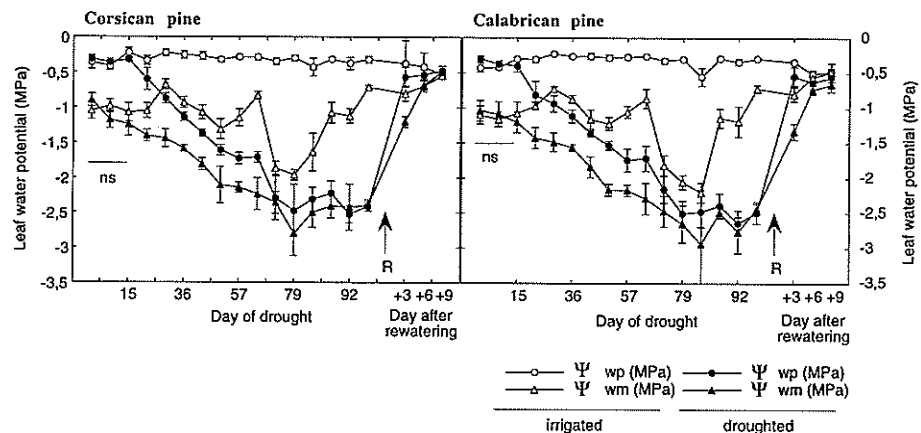


Figure 1. Leaf water potential (Ψ_{wp} and Ψ_{wm} , MPa) for both treatments and varieties in relation to drought. Vertical bars indicate \pm sd. ns = no significant ($P < 0.05$). Data points are the means of four plants within each treatment and variety. R and black arrow indicate the onset of rewatering.

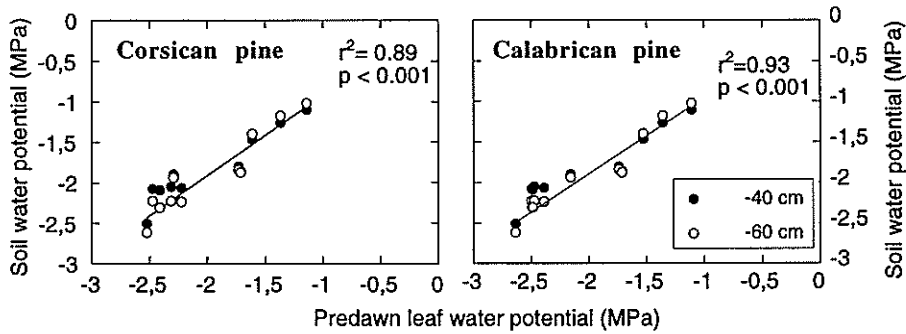


Figure 2. Predawn leaf water potential (Ψ_{wp} , MPa) in droughted plots in relation to soil water potential (MPa). First value corresponds to the 36th day without water.

3.2. Stomatal conductance and net CO_2 assimilation rate

As illustrated in figure 3, under water stress stomatal conductance (g_w) decreased rapidly in both varieties. The decrease in photosynthesis (A) occurred later when g_w presented a decrease of about 50 % of the initial values (below $30 \text{ mmol m}^{-2} \text{ s}^{-1}$) (figure 4). For both varieties, g_w and A stabilized around minimal values of $11 \text{ mmol m}^{-2} \text{ s}^{-1}$ and $0.5 \mu\text{mol m}^{-2} \text{ s}^{-1}$ after 43 days of drought. Rewatering induced a rapid recovery of stomatal conductance and CO_2 assimilation.

In watered treatments, g_w and A showed considerable variability due to plant-to-plant variability and weather fluctuation during the season. Through the season, the average values were $76.9 \text{ mmol m}^{-2} \text{ s}^{-1}$ and $2.9 \mu\text{mol m}^{-2} \text{ s}^{-1}$ for Calabrian pine and $79.0 \text{ mmol m}^{-2} \text{ s}^{-1}$ and $3.6 \mu\text{mol m}^{-2} \text{ s}^{-1}$ for Corsican pine for g_w and A , respectively (figure 4).

Both varieties showed a similar evolution of A and g_w in response to decreasing Ψ_{wp} (figure 5). Stomatal conductance decreased sharply between -0.4 and -1.1 MPa. Inhibition of A started below -1.1 MPa but dropped rapidly thereafter. g_w

and A reached values near zero when Ψ_{wp} reached -1.6 MPa.

3.3. Plant growth and dry matter

In 1992, no mortality or visible damage was observed at the end of the first growing season. However, the first season terminal shoot growth was significantly lower for Calabrian pine than for Corsican pine (table II).

In both varieties, water stress during the second growing season had no influence on annual stem elongation, but reduced significantly stem diameter (mean value for both varieties: -20 %) and length of new needles (mean value for both varieties: -25 %) (table III). Under well-watered conditions, a significant difference was also noted on needle length which appeared shorter in Calabrian pine than in Corsican pine (-12 %) (table III). That was expected because needle length is one of the morphological differences between these two varieties [18].

In both varieties, total elaborated dry weight in the dry regime averaged less than 60 g, while elaborated biomass in the irrigated plots averaged over 95 g (table IV). Drought was responsible for a

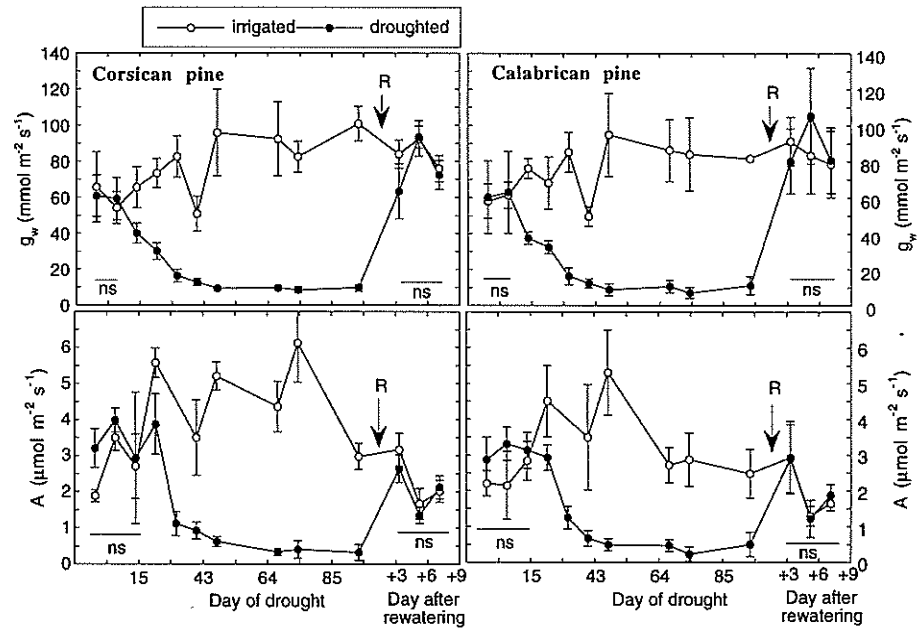


Figure 3. Net assimilation rate per unit leaf area (\pm sd) (A , $\mu\text{mol m}^{-2} \text{s}^{-1}$) and leaf stomatal conductance to water vapour (\pm sd) (g_w , $\text{mmol m}^{-2} \text{s}^{-1}$) in relation to drought. Verticals bars indicate (\pm sd). ns = no significant ($P < 0.05$). Data points are the means of four plants within each treatment and variety. R and black arrow indicate the onset of rewatering.

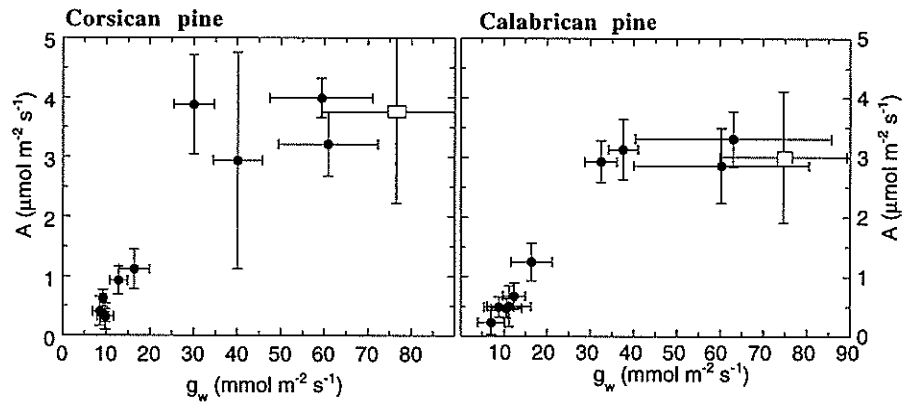


Figure 4. Relationship between net assimilation rate per unit leaf area (\pm sd) (A , $\mu\text{mol m}^{-2} \text{s}^{-1}$) and leaf stomatal conductance to water vapour (\pm sd) (g_w , $\text{mmol m}^{-2} \text{s}^{-1}$) in droughted plots. Data points are the means of four plants. White rectangle indicates the mean value (\pm sd) of irrigated plants over the season.

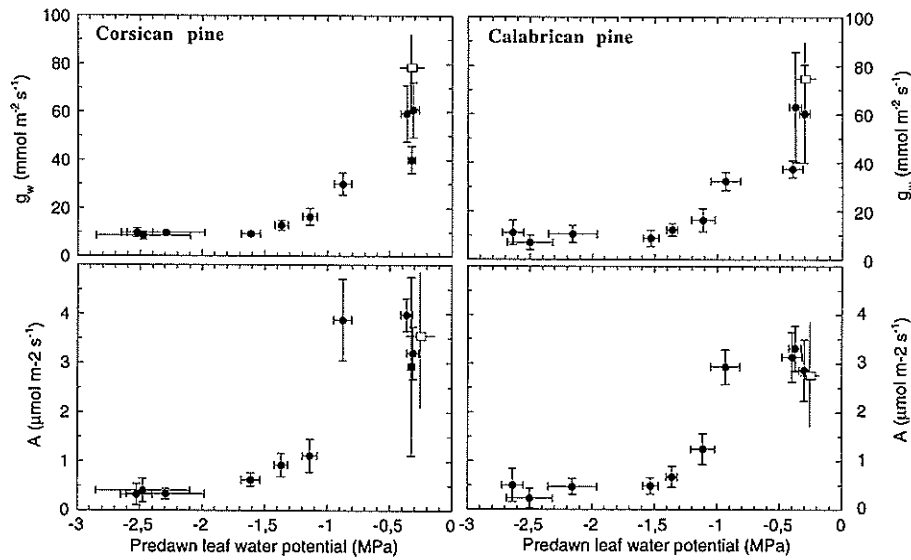


Figure 5. Net assimilation rate per unit leaf area (\pm sd) (A , $\mu\text{mol m}^{-2} \text{s}^{-1}$) and leaf stomatal conductance to water vapour (\pm sd) (g_w , $\text{mmol m}^{-2} \text{s}^{-1}$) in relation to predawn leaf water potential (\pm sd) (Ψ_{wp} , MPa) in droughted plots. Data points are the means of four plants. White rectangle indicates the mean value (\pm sd) of irrigated plants over the season.

Table II. Seedling size at the end of the first growing season. Means followed by the same letter are not significantly different at the $P < 0.05$ level.

	Calabrian pine ($n = 140$)	Corsican pine ($n = 125$)
Total height (cm)	19.4 ± 4 a	21.2 ± 5.3 b
Annual stem elongation (cm)	8.7 ± 3.1 a	11.0 ± 3.7 b

Table III. Seedling size at the end of the second growing season in the different treatments. For a given parameter and variety, mean values not sharing common letters are significantly different at the $P < 0.05$ level.

	Calabrian pine		Corsican pine	
	Irrigated ($n = 68$)	Droughted ($n = 69$)	Irrigated ($n = 62$)	Droughted ($n = 61$)
Total height (cm)	46.3 ± 13 a	48.8 ± 11.3 a	50.1 ± 12.6 a	48.3 ± 14.2 a
Annual stem elongation (cm)	26.1 ± 10.3 a	29.9 ± 8.5 a	27.9 ± 9.9 a	27.9 ± 8.8 a
Annual stem diameter (mm)	9.4 ± 2.9 a	7.8 ± 1.6 b	9.5 ± 2.7 a	7.1 ± 2 b
Needles length (cm)	13 ± 2 a	10 ± 1.4 b	14.8 ± 2 c	10.7 ± 2 b
($n =$ number of sampled needles)	$n = 558$	$n = 618$	$n = 580$	$n = 476$

Table IV. Dry weight partitioning of the plants at the end of the second growing season in the different treatments. For a given parameter and variety, mean values not sharing common letters are significantly different at the $P < 0.05$ level.

	Calabrian pine		Corsican pine	
	Irrigated ($n = 14$)	Droughted ($n = 19$)	Irrigated ($n = 13$)	Droughted ($n = 13$)
Stem dry weight (g)	29.4 ± 15.7 a	13.5 ± 6.4 b	29.4 ± 10.9 a	15.5 ± 8.2 b
Leaf dry weight (g)	43.1 ± 18.1 a	26.4 ± 10.3 b	46.8 ± 16.8 a	32.1 ± 15.0 b
Root dry weight (g)	23.7 ± 12.4 a	10.3 ± 4.4 b	22.0 ± 10.3 a	10.1 ± 4.9 b
Total dry weight (g)	96.2 ± 44.2 a	50.2 ± 20.4 b	98.2 ± 36.2 a	57.8 ± 26.9 b
SMR ($\text{g}\cdot\text{g}^{-1}$)	0.30 ± 0.04 a	0.26 ± 0.04 b	0.30 ± 0.02 a	0.26 ± 0.04 b
LMR ($\text{g}\cdot\text{g}^{-1}$)	0.46 ± 0.06 a	0.53 ± 0.04 b	0.48 ± 0.03 a	0.56 ± 0.05 b
RMR ($\text{g}\cdot\text{g}^{-1}$)	0.24 ± 0.05 a	0.20 ± 0.03 b	0.21 ± 0.04 b	0.18 ± 0.03 c
R/S ($\text{g}\cdot\text{g}^{-1}$)	0.32 ± 0.08 a	0.26 ± 0.05 bc	0.28 ± 0.07 ab	0.22 ± 0.06 c

decrease of about 51, 36 and 42 % in stem, needle and root elaborated dry matter respectively.

Drought led to a marked decrease in SMR, RMR and R/S ratio and to an increase in LMR (table IV). Moreover, in both treatments, RMR was lower in Corsican pine than in Calabrian pine. At the end of the drying cycle, no massive leaf abscission was observed.

4. DISCUSSION AND CONCLUSIONS

Three-year-old Calabrian and Corsican pines exhibited similar decrease of net CO_2 assimilation rate and stomatal conductance with increasing drought. Our results are in agreement with previous work on *Pinus nigra* [4–7, 32] and confirm the water stress adaptation of this collective Mediterranean species which appears to be linked to the ability to avoid internal water stress. Similar features have been obtained in a wide range of Pinus and Mediterranean species: *Pinus uncinata* and *Pinus pinaster* [6], *Pinus sylvestris* [45], *Abies bornmuelleriana* [25], *Quercus afares* and *Quercus faginea* [1]. However, it must be noticed that the

Mediterranean habitats are characterized by great climatological and ecological diversities which have induced differential drought adaptations in the different species [40]. Thus, a lot of other Mediterranean species exhibit a drought-tolerance strategy: *Cedrus atlantica*, *Quercus pubescens*, *Quercus ilex*, *Buxus sempervirens*, etc. [20]. Thus, it appears that there is no common Mediterranean strategy to moisture stress response.

As previously observed on Norway spruce [11, 37] and in several oak species [8, 42], care must be taken with the use of predawn water potential as a driving variable of stomatal closure. In our study, a large decrease of g_w (about 30 %) was observed with no obvious variation in predawn water potential ($\Psi_{wp} = -0.4$ MPa) suggesting that this parameter is not always the best precocious indicator of the water stress actually experienced by plants. However, when drought increases, the close correspondence of predawn with soil water potentials supports their use as an indicator of soil moisture. There is still considerable uncertainty over the underlying causes of drought-induced changes in plant feature. However, in our study, we may suggest that the superficial root system of the seedlings has probably

played an important role in the early variations of g_w by precociously detecting the increase in soil hydraulic resistance [14, 39, 47]. Many recent studies ascribe stomatal closure to a single chemical substance, such as cytokinin or abscisic acid, transported from the roots in the drying zones of the soil to the leaves by means of xylem flux [13, 15, 49]. In our study, decrease in plant hydraulic conductance through cavitation and embolism in the xylem conduits [30, 48] seems not to be responsible for the effect of drought on physiological processes, because the minimum value of predawn water potential (-2.5 MPa) remained higher than the threshold of -4 MPa inducing a significant xylem cavitation for these varieties [24].

After the first growing season, height growth differences suggest a more severe transplanting shock for Calabrian pine. The mechanisms underlying these differences remain unclear but may involve root regeneration and elongation decrease over the first planting year [26].

As expected, the summer water stress had a significant impact on growth and severely reduced whole-plant biomass accumulation by a factor of 1.9 in both varieties. The annual stem elongation was not affected by drought because most of the elongation occurred during the spring when water was available in large amounts. Terminal buds started to break in early April and stopped growing by late June, whereas radial increment continued till September [34]. This phase of growth is in agreement with previous results and confirms the monocyclic shoot growth in *Pinus nigra* [27, 29, 33]. The seasonal pattern in root growth has not been investigated in the present study. However, Corsican pine typically has two peaks in root growth. A low activity during spring till the end of June followed by a period of important root growth from July to September [2, 43]. Based on these observations, we may suggest that the water

stress was applied during the maximum root growth period, which could explain why the dry regime reduced belowground growth proportionally more than aboveground growth, causing a significant decrease in the R/S ratio. This response diverges with models of whole plant biomass partitioning that predict that low rates of water absorption will result in increased biomass partitioning to root growth [12, 28, 38, 41]. However, it is clear that partitioning of assimilates between roots and aerial parts may differ under long- and short-term drought and that water stress also has a greater effect during certain phases of the plant's cycle than others [31]. For the root system, although the differences were small and not always statistically different (at $P < 0.05$) (table IV), the data might suggest a greater investment in root growth in Calabrian pine than in Corsican pine. From the perspective of morphological adaptation to drought, further investigations could be undertaken to confirm this trend.

Rewatering rapidly restored stomatal conductance, photosynthesis and water status to well-watered conditions. In addition to a capacity for avoidance of dehydration through stomatal closure, the rapid recovery in gas exchange might also indicate a robust, dehydration-tolerant photosynthetic apparatus. Nevertheless, a significant shoot growth decrease was observed the following year [34], suggesting important after effects of unfavourable climatic conditions on tree growth.

In conclusion, the pine plants were characterized by a high sensitivity to drought associated with an efficient stomatal control of transpirational water loss. With the classical ecophysiological parameters studied, the expression of the genetic characteristics between varieties in drought tolerance appears to be limited. This preliminary study suggests further investigations which could be undertaken to learn

about drought feature at cell and molecular levels (osmotic adjustments, carbohydrate storage, etc.). A better understanding of the physiological basis of the responses to stress will contribute to increasing the efficiency of selection for improved yield performance under stress. Moreover, these results are consistent with dendroecological results and highlight the physiological underlying mechanisms to the long-term growth decrease observed in mature stands in response to water stress.

ACKNOWLEDGMENTS

The authors would like to thank J.M. Guehl for helpful discussion during the preparation of this article and two anonymous reviewers for their comments and for the critical revision of the manuscript. The authors also express their gratitude to Daniel Himbert and Yves Lefevre for technical assistance.

REFERENCES

- [1] Acherar M., Rambal S., Lepart J., Evolution du potentiel hydrique foliaire et de la conductance stomatique de quatre chênes méditerranéens lors d'une période de dessèchement, *Ann. Sci. For.* 48(5) (1991) 561-573.
- [2] Arbez M., Croissance des racines du Pin laricio de Corse au stade juvénile. Relation avec la croissance des parties aériennes et les facteurs du milieu, *Ann. Sci. For.* 28(3) (1971) 259-288.
- [3] Arbez M., Miller C., Contribution à l'étude de la variabilité géographique du *Pinus nigra* Arnold, *Ann. Sci. For.* 28(1) (1971) 23-49.
- [4] Aussenac G., Comportement hydrique de rameaux excisés de quelques espèces de sapins et de pins noirs en phase de dessiccation, *Ann. Sci. For.* 37(3) (1980) 201-215.
- [5] Aussenac G., Granier A., Quelques résultats de cinétique journalière du potentiel de sève chez les arbres forestiers, *Ann. Sci. For.* 35(1) (1978) 19-32.
- [6] Aussenac G., Valette J.C., Comportement hydrique estival de *Cedrus atlantica* Manetti, *Quercus ilex* L., *Quercus pubescens* Willd. et de divers pins dans le Mont Ventoux, *Ann. Sci. For.* 39(1) (1982) 41-62.
- [7] Becker M., Transpiration et comportement vis-à-vis de la sécheresse de jeunes plants forestiers (*Abies alba* Mill., *Picea abies* L. Karsten., *Pinus nigra* Am. ssp. *laricio* Poirét., *Pinus strobus* L.), *Ann. Sci. For.* 27(4) (1970) 401-420.
- [8] Bréda N., Cochard H., Dreyer E., Granier A., Seasonal evolution of water transfert in a mature oak stand (*Quercus petraea* (Matt.) Liebl) submitted to drought, *Can. J. For. Res.* 23 (1993) 1136-1143.
- [9] Caemmerer S., Farquhar G.D., Some relationships between the biochemistry of photosynthesis and the gas exchange of leaves, *Planta* 153 (1981) 376-387.
- [10] Christensen K.I., Comments on the earliest validly published varietal name for Corsican pine, *Taxon* 42 (1993) 649-653.
- [11] Cienciala E., Lindroth A., Cermak J., Hällgren J.E., Kucera J., The effects of water availability on transpiration, water potential and growth of *Picea abies* during a growing season, *J. Hydrol.* 155 (1994) 55-71.
- [12] Comeau P.G., Kimmins J.P., Above and below-ground biomass and production of lodgepole pine on sites with differing soil moisture regimes, *Can. J. For. Res.* 19 (1989) 447-454.
- [13] Davies W.J., Jones H.G., Abscisic acid. Physiology and biochemistry, in: Davies W.J., Jones H.G. (Eds.), *Environmental Plant Biology Series*, 1991, 261 p.
- [14] Davies W.J., Metcalfe J., Lodge T.A., Da Costa A.R., Plant growth substances and regulation of growth under drought, *Aust. J. Plant. Physiol.* 13 (1986) 105-125.
- [15] Davies W.J., Tardieu F., Trejo C.L., How do chemical signals work in plants that grow in drying soil? *Plant Physiol.* 104 (1994) 309-314.
- [16] Debazac E.F., Le pin laricio de Corse dans son aire naturelle, *Rev. For. Fr.* 16(3) (1964) 188-215.
- [17] Debazac E.F., Les pineraies de Calabre et de Sicile, *Rev. For. Fr.* 17(10) (1965) 662-673.
- [18] Debazac E.F., Les modalités de la croissance en longueur chez les pins. Bulletin de la Société Botanique de France, Mémoire 1966, Colloque sur la physiologie de l'arbre, pp. 3-14.
- [19] Debazac E.F. Contribution à la connaissance de la répartition et de l'écologie de *Pinus nigra* Arnold dans le sud-est de l'Europe, *Ann. Sci. For.* 28(2) (1971) 91-139.
- [20] Ducrey M., Réactions à la sécheresse de quelques espèces forestières méditerranéennes, *Rev. For. Fr.* 40(5) (1988) 359-380.
- [21] Fineschi S., Grossoni P., Contenuto in monoterpeni di oleoresine xilematiche in

- provenienze di pino laricio, Italia Forestate e Montana 36(5) (1981) 232-239.
- [22] Gathy P., Étude de quelques caractères de forme de diverses races de *Pinus nigra* Arnold, Bull. Soc. R. For. Belg. 68(12) (1961) 557-569.
- [23] Gilbert J.M., Chevalier R., Dumas Y., Autécologie du pin laricio de Corse dans le secteur ligérien, Rev. For. Fr. 48(3) (1996) 201-216.
- [24] Girard S., Clement A., Boulet-Gercourt B., Cochard H., Guehl J.M., Effects of desiccation on post-planting stress in bare-root Corsican pine seedlings, Tree Physiol. 17(7) (1997) 429-435.
- [25] Guehl J.M., Aussenac G., Bouachrine J., Zimmermann R., Pennes J., Ferhi A., Grieu P., Sensitivity of leaf exchange to atmospheric drought soil drought and water use efficiency in some Mediterranean Abies species, Can. J. For. Res. 21 (1991) 1507-1515.
- [26] Guehl J.M., Clement A., Kaushal P., Aussenac G., Planting stress, water status and non-structural carbohydrate concentrations in Corsican pine seedlings, Tree Physiol. 12 (1993) 173-183.
- [27] Guyon J.P., Betored B., Kremer A., Influence de la température et des précipitations sur l'activité méristématique apical dans une régénération naturelle de pin noir d'Autriche (*Pinus nigra* Arn. ssp. *nigricans* Host), Acta Oecologica Ecol. Plant 8(22) 4 (1987) 309-320.
- [28] Hunt R., Lloyd P.S., Growth and partitioning, New Phytol. 106 (1987) 235-249.
- [29] Ishik K., Seasonal course of height and needle growth in *Pinus nigra* grown in summer-dry Central Anatolia, For. Ecol. Manag. 35 (1990) 261-270.
- [30] Jones H.C., Sutherland R.A., Stomatal control of xylem embolism, Plant, Cell Environ. 14 (1991) 607-612.
- [31] Jones H.G., Flowers T.J., Jones M.B., Plants under stress, in: Jones H.G., Flowers T.J., Jones M.B. (Eds.), Biochemistry, Physiology and Ecology and their Application to Plant Improvement, Cambridge University Press, 1989, 249 p.
- [32] Kaushal P., Aussenac G., Drought preconditioning of Corsican Pine and Cedar of Atlas seedlings, photosynthesis, transpiration and root regeneration after transplanting, Acta Oecologica 11(1) (1990) 61-78.
- [33] Lanner R.M., Patterns of shoot development in *Pinus* and their relationship to growth potential, in: Cannell M.G.R., Last F.T. (Eds.), Tree Physiology and Yield Improvement, Academic Press, London, 1976, pp.223-243.
- [34] Lebourgeois F., Étude dendroécologique et écophysiological du pin laricio de Corse (*Pinus nigra* Arnold ssp. *laricio* Poiré var. *corsicana*) en région Pays de la Loire, Thèse de l'Université de Paris-Sud Orsay en Sciences de la Vie (1995) 209 p.
- [35] Lebourgeois F., Becker M., Dendroécologie du pin laricio de Corse dans l'Ouest de la France. Evolution du potentiel de croissance au cours des dernières décennies, Ann. Sci. For. 53(5) (1996) 931-946.
- [36] Lebourgeois F., Levy G., Becker M., Lefevre Y., Rôle de la nutrition minérale et de l'approvisionnement en eau sur la croissance du pin laricio de Corse dans l'ouest de la France, Ann. Sci. For. 54 (3) (1997) 279-300.
- [37] Lu P., Biron P., Bréda N., Granier A., Water relations of adult Norway spruce (*Picea abies* (L.) Karst) under soil drought in the Vosges mountains: water potential, stomatal conductance and transpiration, Ann. Sci. For. 52 (1995) 117-129.
- [38] McMillin J.D., Wagner M.R., Effects of water stress on biomass partitioning of Ponderosa Pine seedlings during primary root growth and shoot growth periods, For. Sci. 41(3) (1995) 594-610.
- [39] Pallardy S.G., Rhoads J.L., Morphological adaptations to drought in seedlings of deciduous angiosperms, Can. J. For. Res. 23 (1993) 1766-1774.
- [40] Pereira J.S., Chaves M.M., Plant water deficits in Mediterranean ecosystems, in: Smith J.A.C., Griffiths H. (Eds.), Water Deficits. Plant Responses from Cell Community, Environmental Plant Biology Series, 1993, pp. 237-251.
- [41] Reader R.J., Jalili A., Grime J.P., Spencer R.E., Matthews N., A comparative study of plasticity in seedling rooting depth in drying soil, J. Ecol. 81 (1993) 543-550.
- [42] Reich P.B., Hinckley T.M., Influence of predawn water potential and soil-to-leaf hydraulic conductance on maximum daily leaf diffusive conductance in two oak species, Funct. Ecol. 3 (1989) 719-726.
- [43] Riedacker A., Arbez M., Croissance et régénération des racines et semis de pins laricio et de pins noirs en chambre climatisée et in situ, Ann. Sci. For. 40(1) (1983) 79-110.
- [44] Running F.W., Environmental and physiological control of water flux through *Pinus contorta*, Can. J. For. Res. 10 (1980) 82-91.
- [45] Rutter A.J., Studies of the water relations of *Pinus sylvestris* in plantation conditions. Responses to variation in soil water conditions, J. Appl. Ecol. 4 (1967) 73-81.
- [46] Scholander P.F., Hammel H.T., Bradstreet E.D., Hemmingsen E.A., Sap pressure in vascular plants, Science 148 (3668) (1965) 339-346.

- [47] Schurr U., Gollan T., Composition of xylem sap of plants experiencing root water stress, a descriptive study, in: Davies W.J., Jeffcoat B. (Eds.), Importance of Root to Shoot Communication in the Responses to Environmental Stresses, BSPGR Monograph 21, 1990, pp. 201–214.
- [48] Sperry J.S., Tyree M.T., Water-stress-induced xylem embolism in three species of conifers, *Plant, Cell Environ.* 13 (1990) 427–436.
- [49] Tardieu F., Davies W.J., Root-shoot communication and whole-plant regulation of water flux, in Smith J.A.C., Griffiths H. (Eds.), Water Deficits. Plant Responses from Cell to Community, Environmental Plant Biology Series, 1993, pp. 147–162.
- [50] Wright J.W., Bull W.I., Geographic variation in European black pine - two year results, *For. Sci.* 8 (1962) 32–42.

1

2