



Root hydraulic conductance, gas exchange and leaf water potential in seedlings of *Pistacia lentiscus* L. and *Quercus suber* L. grown under different fertilization and light regimes

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ABSTRACT

Differences in morphology, biomass allocations and physiological responses were investigated in seedlings of Mastic tree (*Pistacia lentiscus* L.) and Cork oak (*Quercus suber* L.) submitted to contrasting fertilization and light regimes during early growth. These species are two evergreen sclerophyllous Mediterranean species frequently used in Mediterranean reforestation programmes. Fertilization was the treatment that affected most of the morphological and physiological variables evaluated in *P. lentiscus* and *Q. suber* seedlings. Leaf area and specific leaf area (SLA) were affected by shading treatment in both species, showing higher values in seedlings grown under shade. *P. lentiscus* seedlings showed a high capacity to modify root morphological variables and root hydraulic conductance (K_R) with the fertilization treatment. In contrast, *Q. suber* showed low to moderate root system changes with the treatments applied, although the fertilization level affected biomass allocation (i.e., root to shoot ratio) in both species. Under high water demand, *P. lentiscus* seedlings with high K_R allowed transpiration (E) to increase without increasing the water potential gradient between soil and leaves. In *Q. suber*, high fertilization induced significant increases in photosynthesis (A), as well as a tendency to increase E with significantly lower leaf water potential (ψ_L).

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1. Introduction

Water stress is one of the main environmental limitations for plants in Mediterranean climate areas (Di Castri, 1981). A long dry season with negligible rainfall, high radiation and high evaporative demand imposes severe stress on plants. These abiotic conditions lead to water deficits that affect many physiological processes and can have consequences for survival and plant growth (Larcher, 2001; Lo Gullo et al., 2003). Species tend to adapt to environmental conditions by different morphological and physiological adjustments (Larcher, 2001; Wood, 2005). This fact is especially important at the seedling stage since seedling establishment and growth in field conditions strongly depend on soil water availability (Vallejo et al., 2000). The capacity of the different species to avoid damaging effects determines their chances of survival and growth (Filella et al., 1998; Vilagrosa et al., 2003).

One of the main morpho-functional traits of species is to optimize water absorption and transport to leaves, thus maximizing their hydraulic system, and this is important when interpreting

leaf physiological behaviour (Tyree et al., 1991; Maherali et al., 1997; Cochard et al., 2002; Bacelar et al., 2007). Water flow through leaves has significant implications for whole plant hydraulics, plant growth, leaf structure, function and ecology. Therefore, limiting the water loss by leaves to a minimum in order to avoid the interruption of water flow in the xylem under conditions of severe drought is of utmost importance (Burghardt and Riederer, 2003). When the threshold is exceeded xylem cavitation occurs and both the growth and survival of the species become compromised (Sperry, 2000; Cochard, 2006). Stomata response has been found to be related to changes in plant hydraulic conductance to prevent desiccation by maintaining the xylem water potential above the minimum threshold (Cochard et al., 2000; Sperry et al., 2002; Vilagrosa et al., 2003). Because the hydraulic conductance determines how much xylem water potential falls below the soil water potential during transpiration, it indicates that there should be a link between the plant's hydraulic traits and the transpirational demand of its foliage (Sperry et al., 1998; Brodrribb and Field, 2000). Moreover, reductions in stomatal aperture to avoid excessive water losses can increase water use efficiency at leaf level, playing an important role in plant response to drought (Cochard et al., 2004; Agele et al., 2005).

In addition to water availability, light and nutrients are two of the most important resources for plant life, and they often interact

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in natural environments (Hirose and Bazzaz, 1998; Lone and Khan, 2007). Soil nutrient availability and light conditions influence the morpho-functional and biomass allocation patterns of development in plants. Previous studies have reported changes in plant morphology and physiological traits as a consequence of different nutrient and light conditions (Villar-Salvador et al., 2004; Barigah et al., 2006; Trubat et al., 2006). Seedlings grown with high fertilization doses have shown high survival and growth under field conditions (Villar-Salvador et al., 2004; Oliet et al., 2009), but these results have been different in environments with strong water limitations (Trubat et al., 2008). Indeed, it has been hypothesized that high-fertilized trees may be more vulnerable to drought stress than low-fertilized ones because of the former's increased aerial size and transpiration rates and decreased allocation to roots (Harvey and Van den Driessche, 1999).

The light environment may also profoundly affect the morphological and physiological characteristics of plants, as it is mainly related to biomass allocation patterns and root system development (Valladares and Pearcy, 1998; Clearwater and Meinzer, 2001). Shading by means of nursery techniques or by means of treeshelters under field conditions has been reported to improve the survival and field establishment of Mediterranean species (Valladares et al., 2000; Sack and Grubb, 2003; Pons and Pausas, 2006; Pérez-Devesa et al., 2008).

The objective of the present study was to analyze the combined effects of nutrient availability and incoming radiation during early growth in two co-occurring Mediterranean sclerophyllous species on the basis of several morphological and physiological traits. Mastic tree (*Pistacia lentiscus* L.) and Cork oak (*Quercus suber* L.) were selected because these species are both common in Mediterranean flora and widely used in reforestation programmes to recover degraded ecosystems. Fertilization and light regimes are known to be important management options in nursery production as their ability to change plant morphological and physiological characteristics can affect seedling performance and drought resistance. We hypothesized that seedlings growing under more stressful conditions (lower nutrients and high light availability) would have a higher capacity to supply water to leaves since they would allocate more resources to belowground than to aboveground parts and, consequently, these seedlings should be able to maintain a better performance.

2. Materials and methods

2.1. Growth conditions

P. lentiscus and *Q. suber* seeds from local provenance were sown in a public nursery (Santa Faz, Alicante, Spain; 38°23'N; 0°26'W; 80 m a.s.l.) in January 2005. The mean annual rainfall is 353 mm and the mean annual temperature is 18 °C. Seedlings were grown for 6 months in 300 cm³ commercial containers (ForestPot®) in a mixture of peat and cocopeat fibre (1:1, v/v). We used a complete factorial design with two factors (nutrient availability and light conditions), each having two levels. The irradiance regimes were full sunlight (FS) and 60% full sunlight (S), as excessive shade is not recommended for Mediterranean plants. Incoming light was manipulated by protecting half of the seedlings with a shade mesh. The photosynthetically active radiation was measured with a Sunfelck Ceptometer (Decagon Devices Inc., Pulman, WA, USA) to determine the incoming radiation received by the seedlings. Around 80 fertilized seedlings per species and treatment were arranged randomly under the different light levels. Nutrient availability was established by applying a slow-release fertilizer (Osmocote®; N:P:K 14:8:14) mixed with the substrate at 2 g l⁻¹ (low fertilization, LF) or 5 g l⁻¹ (high fertilization, HF)

doses. The watering regime was gauged to the seedling water demand.

2.2. Morphological traits

After 6 months, 7 seedlings from each treatment and species were randomly selected for morphological variables determination. Stem height and root collar diameter were measured. Leaves were scanned and leaf area was measured using image analysis software (WinRhizo, Régent Instruments Inc., Quebec, Canada). Specific leaf area (SLA) was calculated as leaf blade area/dry blade mass. Root surface area and root length were measured after washing out the roots, scanning them (on a professional scanner with transparency adapter) and analyzing the images with the same specific analysis software used for the leaves. Biomass fractions of leaves, stems and roots were determined after drying in a forced-air oven at 65 °C to constant weight. Biomass allocation at the plant level was determined between roots and aboveground plant parts, commonly expressed as the root to shoot ratio on a dry weight basis (Cairns et al., 1997).

2.3. Root hydraulic conductance

Whole root system hydraulic conductance (K_R) was measured in 7 plants per treatment in the laboratory. Seedlings were watered to field capacity the night before the measurements. K_R measurements were carried out early in the morning. Root systems were kept in their substrate and perfused with distilled and de-gassed ultra-pure water filtered through a 0.1 µm water filtration membrane using the high pressure flow meter method (HPFM, Dynamax, USA) connected to the stump. K_R was measured in a transient way within a range of 0–0.5 MPa pressures at a constant rate of 3–5 kPa s⁻¹ and the flow rate (F) was recorded every 3 s as described in Tyree et al. (1995). F was plotted versus pressure (P), and K_R was estimated as the slope of a linear regression between both variables (ΔF , ΔP). K_R was corrected for air temperature. Laboratory air temperature was registered in each measurement and varied no more than 1.5 °C during any set of measurements. Substrate temperature was assumed to be the same as air temperature. We estimated root hydraulic conductance on a leaf area basis (K_{RL}), root hydraulic conductance scaled by total root surface area (K_{RR}), and root hydraulic conductance scaled by root length (K_{RRL}) (Nardini et al., 2000).

2.4. Gas exchange and leaf water potential

Gas exchange measurements were taken in 5 well-irrigated seedlings per treatment and species (a total of 20 seedlings per species) in a climatic chamber Fytotron (Sanyo, Gallenkamp PLC, Leicester). Environmental conditions in the climatic chamber were adjusted so as to submit seedlings to highly stressful conditions. Maximum summer air temperatures of 35–40 °C frequently occur in the Mediterranean maquis (Larcher, 2000). Seedlings were subjected to a photoperiod of 12 h in the climatic chamber (temperature: 35 °C day/25 °C night; relative humidity: 44% day/55% night), with an ambient vapour pressure deficit of 3.15 ± 0.10 kPa during day conditions and a photosynthetic active radiation of 1000 µmol/m² s. Net photosynthesis (A) and transpiration rate (E) were measured at the beginning of the daytime climatic conditions and consecutively in all plants until constant values (about 3 h later). Gas exchange measurements were made at 380 ppm of CO₂ concentration and 1000 µmol/m² s of photosynthetic active radiation by using a portable infrared gas analyzer Li-6400 (Li-COR Biosciences, UK Ltd.). Instantaneous water use efficiency (WUE) was estimated as the ratio between A and E , µmol CO₂ fixed per mmol H₂O used per unit of leaf area and unit of time (Larcher, 2001). When the gas exchange measurements registered in each

seedling were constant, leaf water potential (Ψ_L) was measured with a Scholander-type pressure chamber in the same seedlings.

2.5. Statistical analysis

Treatment effects on seedlings in the studied variables (morphology, biomass allocation, root hydraulic conductance, gas exchange and leaf water potential) for both species were analysed by two-way analysis of variance (ANOVA), with fertilization and light as fixed factors. The analyses were made for each of the species studied. When it was necessary the data were transformed to assure ANOVA assumptions. All statistical analyses were performed using the SPSS 15.0 statistical package (SPSS Inc., Chicago, USA).

3. Results

3.1. Morphological and biomass allocation traits

Fertilization produced more significant changes in morphology and biomass allocation than shading in both species. In *P. lentiscus*, stem height showed significant differences in response to fertilization and light treatments, with highest values in seedlings grown under high fertilization and shading treatments (Tables 1 and 2). Despite changes in stem height, no differences in the root collar diameter of *P. lentiscus* seedlings were found among the four treatments. The high fertilization treatment promoted the highest stem and root collar diameter development in *Q. suber*, while there were no differences in the response to light treatments (Tables 1 and 2).

Shading and high fertilization significantly increased the leaf area in both species. Specific leaf area (SLA) was significantly influenced only by the light treatment in the two species, increasing in seedlings grown under shade conditions. Root surface area and root length values were statistically higher in the high fertilization treatment in *P. lentiscus*, but these variables were not affected by either the fertilization or the light treatment in *Q. suber* (Tables 1 and 2).

Leaf and stem biomass increased in response to the high fertilization treatment in both species but no differences were observed in response to the light treatment. Root biomass significantly

increased in the high fertilization treatment in *P. lentiscus*, showing the highest values in the seedlings growing under full sunlight and high fertilization conditions. However, in *Q. suber* root biomass was not affected by the applied treatments. Both species significantly decreased their root to shoot ratio in response to the high fertilization treatment (Tables 1 and 2).

3.2. Root hydraulic conductance, gas exchange and leaf water potential

P. lentiscus responded to differences in nutrient availability, showing a higher capacity to modify root hydraulic conductance (K_R) than *Q. suber*. Significant differences in leaf specific root hydraulic conductance (K_{RL}) and root hydraulic conductance by root surface area (K_{RR}) were found with the fertilization treatment in *P. lentiscus* (Fig. 1, Table 3). *P. lentiscus* seedlings grown under high fertilization showed lower values of K_{RL} and K_{RR} than those grown under low fertilization doses, with an average reduction of 47–67% in response to high fertilization (Fig. 1). Root hydraulic conductance scaled by root length (K_{RRL}) showed a tendency to decrease with high fertilization doses in *P. lentiscus*, but this decrease was not statistically significant (Table 3). No significant differences in K_{RL} , K_{RR} and K_{RRL} were measured in *Q. suber* seedlings in response to the fertilization and light treatments (Fig. 1, Table 3).

Net photosynthesis (A) measured under high vapour pressure deficit significantly increased in *Q. suber* in response to high fertilization in both full sunlight and shade, but no significant differences were observed in *P. lentiscus* (Fig. 2, Table 3). However, transpiration (E) was significantly lower in the high fertilization treatment in *P. lentiscus* seedlings grown under full sunlight and shade (Fig. 2, Table 3). As a consequence, water use efficiency (WUE) showed significant differences with the fertilization treatment in *P. lentiscus*. WUE was higher (by an average of 23%) in seedlings grown in high fertilization than in low fertilization treatments without the influence of light conditions. No significant effect of the treatments in WUE was observed in *Q. suber* seedlings (Fig. 2, Table 3).

Leaf water potential (Ψ_L) was significantly affected by the light treatment in *P. lentiscus* and by the fertilization treatment in *Q. suber*

Table 1
Morphological variables and biomass traits for *Pistacia lentiscus* and *Quercus suber* seedlings growing under different light and fertilization levels. Values are the means \pm SE ($n = 7$).

Variables	Species	Full sunlight		Shading	
		LF	HF	LF	HF
Stem height (cm)	<i>P. lentiscus</i>	12.06 \pm 0.28	15.16 \pm 1.69	14.59 \pm 0.90	24.29 \pm 1.23
	<i>Q. suber</i>	21.18 \pm 2.18	34.43 \pm 2.65	26.00 \pm 2.59	42.83 \pm 5.26
Root collar diameter (mm)	<i>P. lentiscus</i>	2.84 \pm 0.82	2.44 \pm 0.37	2.50 \pm 0.18	3.56 \pm 0.72
	<i>Q. suber</i>	2.85 \pm 0.18	3.55 \pm 0.98	2.46 \pm 0.20	3.13 \pm 0.14
Leaf area (cm ²)	<i>P. lentiscus</i>	32.21 \pm 4.25	70.18 \pm 12.72	64.13 \pm 6.49	108.63 \pm 17.38
	<i>Q. suber</i>	54.01 \pm 7.14	96.13 \pm 8.94	78.54 \pm 3.67	123.30 \pm 6.12
Specific leaf area (cm ² g ⁻¹)	<i>P. lentiscus</i>	74.88 \pm 2.54	68.75 \pm 6.46	89.67 \pm 4.69	101.58 \pm 11.36
	<i>Q. suber</i>	81.13 \pm 2.51	88.55 \pm 3.04	100.55 \pm 2.99	97.07 \pm 3.06
Root surface area (cm ²)	<i>P. lentiscus</i>	172.87 \pm 41.19	229.08 \pm 30.97	168.69 \pm 16.98	338.36 \pm 45.55
	<i>Q. suber</i>	121.79 \pm 26.29	152.29 \pm 33.90	116.52 \pm 14.67	121.27 \pm 12.96
Root length (m)	<i>P. lentiscus</i>	12.28 \pm 3.42	17.89 \pm 2.94	14.30 \pm 1.07	27.28 \pm 3.85
	<i>Q. suber</i>	10.61 \pm 2.98	11.94 \pm 3.19	10.41 \pm 1.76	11.32 \pm 1.17
Leaf biomass (g)	<i>P. lentiscus</i>	0.55 \pm 0.08	1.64 \pm 0.18	0.85 \pm 0.15	1.55 \pm 0.17
	<i>Q. suber</i>	0.86 \pm 0.11	1.51 \pm 0.15	0.98 \pm 0.11	1.68 \pm 0.22
Stem biomass (g)	<i>P. lentiscus</i>	0.20 \pm 0.04	0.56 \pm 0.05	0.30 \pm 0.04	0.58 \pm 0.10
	<i>Q. suber</i>	0.53 \pm 0.08	1.14 \pm 0.11	0.67 \pm 0.14	1.26 \pm 0.22
Root biomass (g)	<i>P. lentiscus</i>	0.57 \pm 0.08	0.85 \pm 0.09	0.62 \pm 0.08	0.73 \pm 0.09
	<i>Q. suber</i>	2.57 \pm 0.33	3.18 \pm 0.30	2.31 \pm 0.22	2.89 \pm 0.39
Root to shoot ratio (g g ⁻¹)	<i>P. lentiscus</i>	0.76 \pm 0.05	0.39 \pm 0.03	0.54 \pm 0.09	0.34 \pm 0.02
	<i>Q. suber</i>	1.85 \pm 0.22	1.21 \pm 0.11	1.42 \pm 0.13	0.98 \pm 0.11

Table 2
F and P values of a two-way analysis of variance, showing the effects of light, fertilization and their interaction on the morphological and biomass variables measured.

Variables	Species	Light		Fertilization		Light × fertilization	
		F	P	F	P	F	P
Stem height (cm)	<i>P. lentiscus</i>	25.86	<0.001	31.18	<0.001	8.29	0.002
	<i>Q. suber</i>	3.79	0.08	19.58	0.001	0.28	0.61
Root collar diameter (mm)	<i>P. lentiscus</i>	0.44	0.51	0.32	0.58	1.58	0.22
	<i>Q. suber</i>	0.84	0.37	19.99	0.001	0.02	0.91
Leaf area (cm ²)	<i>P. lentiscus</i>	9.45	0.006	12.98	0.002	0.08	0.78
	<i>Q. suber</i>	14.70	0.001	41.50	<0.001	0.04	0.85
Specific leaf area (cm ² g ⁻¹)	<i>P. lentiscus</i>	10.57	0.004	0.16	0.70	1.52	0.23
	<i>Q. suber</i>	23.03	<0.001	0.46	0.51	3.50	0.07
Root surface area (cm ²)	<i>P. lentiscus</i>	2.32	0.14	12.02	0.002	0.00	0.96
	<i>Q. suber</i>	0.59	0.45	0.56	0.46	0.30	0.59
Root length (m)	<i>P. lentiscus</i>	3.75	0.07	9.95	0.005	1.57	0.23
	<i>Q. suber</i>	0.03	0.87	0.21	0.65	0.01	0.93
Leaf biomass (g)	<i>P. lentiscus</i>	0.45	0.50	34.53	<0.001	1.66	0.21
	<i>Q. suber</i>	1.00	0.33	22.35	<0.001	0.07	0.79
Stem biomass (g)	<i>P. lentiscus</i>	1.01	0.32	27.42	<0.001	0.48	0.49
	<i>Q. suber</i>	0.40	0.53	21.64	<0.001	0.27	0.61
Root biomass (g)	<i>P. lentiscus</i>	0.14	0.71	4.96	0.04	1.20	0.28
	<i>Q. suber</i>	0.78	0.39	3.58	0.07	0.00	0.98
Root to shoot ratio (g g ⁻¹)	<i>P. lentiscus</i>	2.97	0.10	25.40	<0.001	0.11	0.75
	<i>Q. suber</i>	3.78	0.06	10.84	0.003	0.25	0.62

Table 3
F and P values of a two-way analysis of variance, showing the effects of light, fertilization and their interaction on the physiological variables measured (root hydraulic conductance, gas exchange and leaf water potential).

Variables	Species	Light		Fertilization		Light × fertilization	
		F	P	F	P	F	P
K_{RL}	<i>P. lentiscus</i>	2.64	0.12	5.68	0.028	2.29	0.15
	<i>Q. suber</i>	2.89	0.10	1.96	0.18	0.54	0.47
K_{RR}	<i>P. lentiscus</i>	0.00	0.97	6.66	0.019	0.03	0.86
	<i>Q. suber</i>	0.73	0.40	0.00	0.99	0.17	0.68
K_{RRL}	<i>P. lentiscus</i>	0.02	0.89	3.95	0.062	0.23	0.64
	<i>Q. suber</i>	0.04	0.84	0.02	0.89	0.46	0.50
A	<i>P. lentiscus</i>	0.04	0.84	0.67	0.42	4.49	0.05
	<i>Q. suber</i>	1.35	0.26	7.92	0.013	0.28	0.61
E	<i>P. lentiscus</i>	0.15	0.71	16.95	0.001	2.85	0.11
	<i>Q. suber</i>	0.93	0.35	3.72	0.07	0.65	0.43
WUE	<i>P. lentiscus</i>	0.78	0.39	38.58	0.001	1.37	0.26
	<i>Q. suber</i>	0.19	0.67	3.23	0.09	0.62	0.44
Ψ_L	<i>P. lentiscus</i>	10.55	0.005	2.76	0.12	0.18	0.93
	<i>Q. suber</i>	1.32	0.27	7.04	0.020	0.61	0.45

(Table 3). In *P. lentiscus*, Ψ_L values were more negative in seedlings grown under shade, while in *Q. suber* the more negative values were found in seedlings submitted to high fertilization treatment (Fig. 3).

4. Discussion

The results of this study showed that fertilization was the treatment that most affected the morphological and physiological variables evaluated in *P. lentiscus* and *Q. suber* seedlings. The negligible effect of the light treatment could be explained by the fact that these species are able to tolerate a certain degree of shading (Maestre et al., 2003; Cardillo and Bernal, 2006; Pérez-Devesa et al., 2008). Other works have reported significant morpho-functional acclimation processes to low light environments in *Quercus* species, but in these studies seedlings were exposed to higher levels of shade (Valladares et al., 2005; Quero et al., 2006, 2008).

Both leaf area and SLA were significantly increased in the seedlings as a consequence of acclimation to shade conditions (Tables 1 and 2). These changes increase light capture efficiency, which is important in Mediterranean evergreen plants submitted to shade (Valladares et al., 2000; Aranda et al., 2005). The increase in leaf area was not proportional to the increase in leaf biomass, indicating that leaves were less sclerophyllous (i.e., higher SLA) under shade than under full sunlight conditions. The lower degree of sclerophylly derived from seedlings exposed to shading conditions could indicate that these seedlings would be less efficient in controlling water losses under drought conditions (Salleo et al., 1997; Corcuera et al., 2002). In fact, *P. lentiscus* showed changes in water potential between shaded and full sunlight seedlings under high evaporative demands not associated with changes in transpiration rates. Passive water losses at leaf level associated with high vapour pressure deficit conditions produced more negative water potentials in *P. lentiscus*. Previous works have already pointed out the harmful effects of drought associated with plants growing under shade (Valladares and Percy, 1998; Aranda et al., 2005).

High fertilization conditions significantly increased the leaf area and the dry mass invested in leaves and stems in both species. *P. lentiscus* also showed significant increases in root surface area, root length and root biomass variables with the fertilization treatment; in *Q. suber*, however, this pattern was not observed. In both species, the increase in shoot biomass was higher than the increase in root biomass, leading to lower root to shoot ratios in plants grown under high nutrient availability. This pattern of response has also been found in other Mediterranean species (Sack and Grubb, 2002; Löf et al., 2005). A high root to shoot ratio at the early stages of development may be an advantage in environments with seasonal drought, like Mediterranean ecosystems (Lloret et al., 1999; Cortina et al., 2008). Therefore, a reduction in the root to shoot ratio together with a decrease in the degree of sclerophylly could restrict water use efficiency and render more fertilized and shaded plants more vulnerable to drought conditions.

The lack of morphological root responses observed in *Q. suber* suggests a conservative pattern of root morphology and development in response to variations in fertilization and light conditions, which is in agreement with the response observed for other *Quercus*

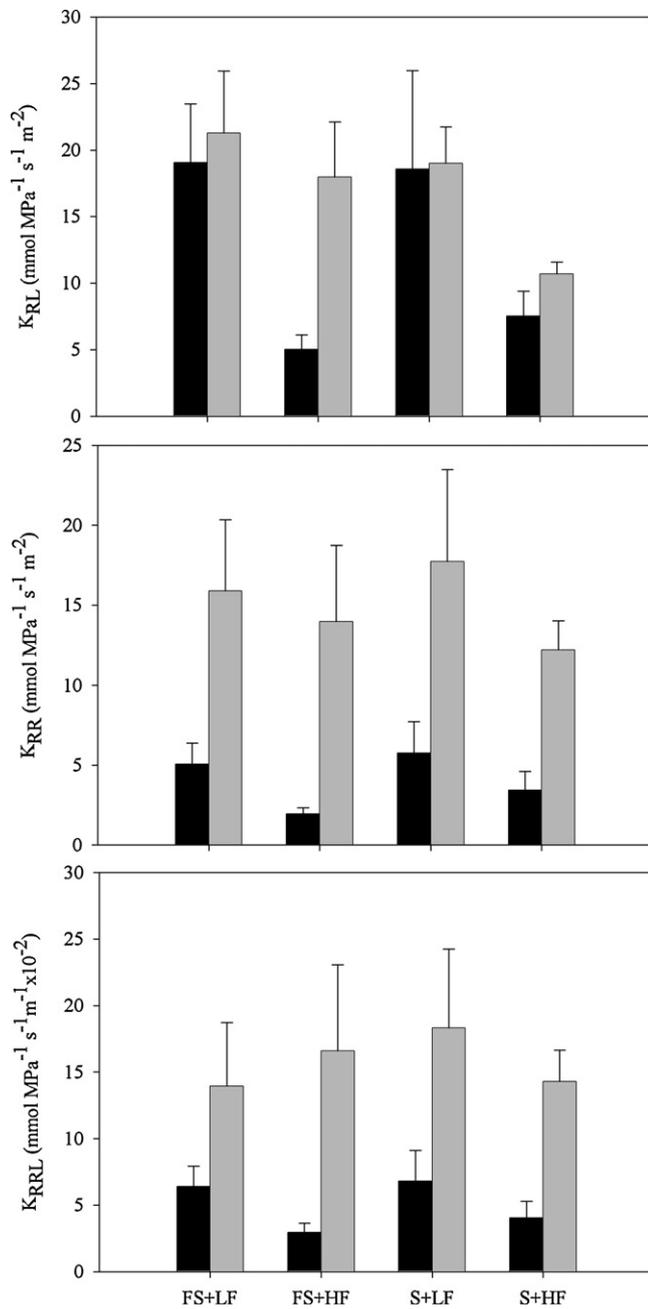


Fig. 1. Leaf specific root hydraulic conductance (K_{RL}), root hydraulic conductance by root surface area (K_{RR}) and root length specific root hydraulic conductance (K_{RRL}) to different light and fertilization availability (FS + LF, full sunlight with low fertilization; FS + HF, full sunlight with high fertilization; S + LF, shade with low fertilization and S + HF, shade with high fertilization) for *P. lentiscus* (black bars) and *Q. suber* (grey bars) seedlings. Data represent the means and error bars are SE, $n = 7$.

species (Berger and Glatzel, 2001; Villar-Salvador et al., 2004; Portsmouth and Niinemets, 2007). However, other studies on *Quercus* species have shown significant alterations in seedling root growth in response to growth conditions (Tsakalimi et al., 2005; Chirino et al., 2008; Cubera et al., 2009). Acorn reserves available during the first stages of seedling development may partially be able to supply the lack of nutrients in the growing medium and thus provide a possible explanation for the results observed in the present work (Welander and Ottosson, 2000; García-Cebrián et al., 2003). Besides the negligible morphological response, K_R in *Q. suber* roots also showed no effect with the fertilization treatment. This lack of any change in root hydraulic conductance in response to

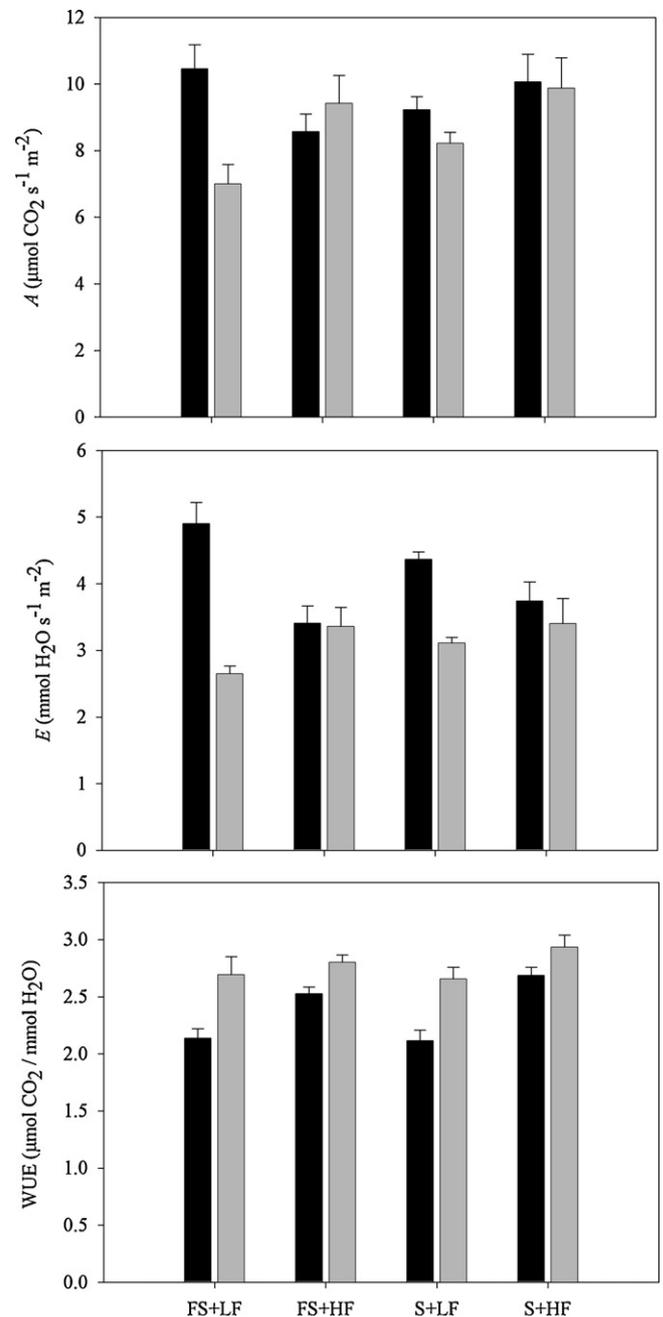


Fig. 2. Net photosynthesis (A), transpiration (E), and water use efficiency (WUE) in all treatments (FS + LF, full sunlight with low fertilization; FS + HF, full sunlight with high fertilization; S + LF, shade with low fertilization and S + HF, shade with high fertilization) of *P. lentiscus* (black bars) and *Q. suber* (grey bars) seedlings under a vapour pressure deficit of 3.15 KPa. Data represent means and error bars are SE, $n = 5$.

the applied treatments could be related to the pattern of root system development in this species. The development of an important taproot as the main strategy in *Q. suber* root development would suggest that the hydraulic characteristics of its root system could be governed by the morphological characteristics of the taproot. In fact, previous works have pointed out changes in hydraulic conductance in *Quercus* sp. root systems in response to changes in the diameter and length of their tap root (Pemán et al., 2006; Chirino et al., 2008). Nevertheless, *Q. suber* seedlings grown under different nutrient availabilities showed different photosynthetic capacities and Ψ_L responses. High fertilization induced significant increases in photosynthesis and a tendency to increase E with significantly lower Ψ_L . Low stomatal conductances could contribute to maintain

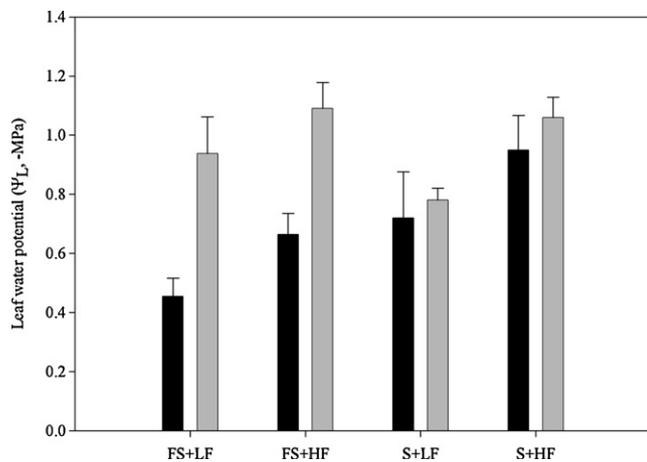


Fig. 3. Leaf water potential (Ψ_L) of *P. lentiscus* (black bars) and *Q. suber* (grey bars) under a vapour pressure deficit of 3.15 kPa. Data represent the means and error bars are SE, $n = 5$.

high leaf water potential, decreasing the risk of xylem cavitation (Mediavilla and Escudero, 2004).

In *Q. suber*, the different adjustments in above- and below ground allocations (i.e., root to shoot ratio) with the high fertilization treatment resulted in a lack of compensation between water loss by E and water supplied by roots, as suggested by the significant decrease in Ψ_L measured. Bucci et al. (2006) reported a decrease in water potential in tree species due to fertilization, suggesting that plants grown with high nutrient availability may be more susceptible to drought stress by means of embolism formation in the xylem.

In *P. lentiscus*, the enhancements in growth caused by high fertilization were not associated with enhanced rates of carbon gain on a leaf area basis under high vapour pressure deficit. Therefore, both a high Rubisco activity and a large concentration of nitrogen in the leaf may be required to achieve the same rate of photosynthesis at lower stomatal conductance (Correia and Diaz-Barradas, 2000; Parry et al., 2005). Stomatal response appeared to be sensitive to changes in K_R in *P. lentiscus*, since E was higher in seedlings with higher K_R (Figs. 1 and 2). In our study, K_R was only affected by the high fertilization treatment in *P. lentiscus* seedlings (Fig. 1, Table 3). There is considerable evidence supporting the link between E and K_R (Nardini et al., 2000; Hubbard et al., 2001; Lo Gullo et al., 2003), since E links the water supply from the roots to the water loss at the leaf surface. High K_R allows E to increase without increasing the water potential gradient between soil and leaves, and this may suggest an important advantage with respect to the drought resistance capacity (Maherali et al., 1997). A high capacity to supply water to leaves may confer drought tolerance by allowing a higher Ψ_L at a given E and soil water supply (Tsuda and Tyree, 2000; Vilagrosa et al., 2003).

Nevertheless, *P. lentiscus* seedlings grown under high fertilization showed higher WUE. A higher WUE has been considered a key factor for providing greater resistance to drought conditions. In addition, many studies have reported the positive influence of nursery fertilization on seedling size and its relation to high seedling survival and growth under field conditions (Puértolas et al., 2003; Villar-Salvador et al., 2004; Luis et al., 2009), although seedling size and survival in water-limited environments has not always been evident (Cortina et al., 1997; Trubat et al., 2008). In this study, *P. lentiscus* seedlings fertilized with low doses showed higher rates of E with similar photosynthetic rates and therefore lower WUE and a less negative water potential gradient than highly fertilized seedlings. On the other hand, seedlings fertilized with high doses showed lower transpiration but the same photosynthetic rates with

higher WUE but more negative water potentials than low-fertilized seedlings. These results from *P. lentiscus* suggest a trade-off between K_R and E in the response to fertilization, a trade-off which influences WUE at leaf level and the water potential gradient at the whole plant level. The relationships found in the present study could influence the survival and growth of this species in water-limited environments. Under high soil water availability, the water potential gradients experienced by high-fertilized seedlings as a consequence of lower K_{RL} would not generate very negative water potentials for these seedlings and they would function above the threshold where seedlings can suffer damage (i.e., xylem cavitation). In this case, high-fertilized seedlings would perform better than low-fertilized seedlings due to a greater carbon gain per unit of water lost (higher WUE). However, under dry soil conditions, more fertilized seedlings would develop more negative water potential values due to lower K_{RL} or they would have to regulate stomatal conductance in order to avoid very negative water potentials with high risk of embolism formation in the xylem (Martínez-Vilalta et al., 2002; Vilagrosa et al., 2003). As a consequence, high-fertilized seedlings would have less carbon gain than low-fertilized seedlings and, thus, the possibility of starving during prolonged periods of time (McDowell et al., 2008).

In conclusion, the light treatment mainly affected morphological leaf traits in both species. The species under study differed in their capacity to acclimate in response to fertilization, with *Q. suber* showing a more rigid pattern of development than *P. lentiscus*, especially at the morpho-functional root level. We conclude that the reduced development of the root system with respect to the shoot system as a consequence of high fertilization could make both species of plants more vulnerable to dieback in the field during drought conditions. In addition, the results obtained in *P. lentiscus* would indicate a trade-off between K_R and E in response to fertilization, influencing WUE at leaf level and the gradient of water potential at the whole plant level. The results reported herein also suggest that the physiological response of *P. lentiscus* seedlings grown under different fertilization regimes could differ depending on the water availability in field conditions.

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